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SELECTION OF PLANTING STOCK FOR VINEYARDS

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Vines can be multiplied by seeds or by buds. Seeds are used only in the origination of new varieties. Commercial vineyards consist of clonal varieties multiplied by buds, the only way in which vines of the desired variety can be obtained.

Growers propagate vines by means of "cuttings." A vine cutting is a segment of mature one-year-old wood including one or more buds. The vine from which the cutting is taken is called the "mother vine" or "parent" and the vine which grows from the cutting, the "daughter vine" or "progeny." Most grape growers and their advisers believe it is necessary for the best results to make a careful selection of the cuttings to be used for propagation. Some act upon this belief and choose a good healthy vineyard noted for the size and quality of its crop, avoiding vines which have borne few or poor grapes.

REASONS FOR SELECTION

The reasons for making this selection are to obtain a daughter vine (1) which will have the desirable qualities of the parent, and (2) which will develop rapidly.

These reasons are based on the beliefs (1) that a bud carries the qualities of the parent, and reproduces them in the offspring, and (2) that the ease and rapidity with which the bud grows depend on its size, maturity, food reserevs and other factors of condition as regards health, vigor and nourishment. While all observers concur in a general way in these beliefs and there is little uncertainty regarding the most favorable conditions for growth and the methods of determining and making use of them, there are some fundamental differences of view regarding the degree to which the characteristics of the parent vine are reproduced in the offspring.

BASES OF SELECTION

We can consider the differences which distinguish two vines as belonging to two classes:

1. Those which distinguish the variety, e.g., the round, black, seedless berries of the Corinth and the elongated, large, white berries of the Olivette. These are heritable or, more properly, transmissible to the offspring by bud or vegetative propagation.

2. Those which lie within the normal range of the variety, e.g., the black color of the Emperor when grown near the coast and the red color of the same grape when grown in the hot interior. These are usually considered as not transmissible. This means that a cutting taken from an Emperor bearing red fruit in Tulare County will produce a vine bearing black fruit if planted in Sonoma County.

The difficulty comes in deciding whether a certain difference belongs to one class or to the other. We have no doubt that the difference between the elongated, asymetrical berry of the Pizzutello and the regular, oblate spheroidal berry of the Palomino is varietal, and therefore transmissible, because, however many berries we examine of either, there is never any doubt to which variety each belongs. This is not the case with the nearly spherical berries which are supposed to characterize the Muscat Gordo Blanco and the obovoid berries which are supposed to characterize the Muscat of Alexandria, because both forms may occur on the same vine. There is doubt, therefore, whether these are different varieties.

Some try to resolve the difficulty by saying that there is only one variety but two "strains" and that if you grow a new vine from a cutting taken from a mother vine which has produced spherical grapes the daughter vine will produce spherical grapes. In other words, the difference observed is transmissible and characteristic of a certain "strain."

However, if the difference is transmissible, it is equivalent to a varietal character and differs only in degree from other undoubted varietal characters.

It is here the whole problem lies—is the difference transmitted by the bud from the mother vine to the daughter vine? In some cases, where there is a distinct difference of kind, this is very easily determined by the growth of offspring. In other cases where there is simply a difference of degree, it is much more difficult. Whether the differences which distinguish the various shades of red exhibited by the Tokay in different localities and even by different vines in the same

vineyard are heritable or not can be determined only by long and careful investigation and well controlled experiments. The reason of this is that the character we are dealing with is greatly influenced by environmental factors such as soil, water and climate.

The most important case and perhaps the most difficult to decide is the "heritability" or transmissibility of degrees of productiveness. To obtain a heavy bearing Muscat vine, are we assisted at all by taking our cuttings from a Muscat mother vine which has borne heavy crops? A belief that we are is the basis of a large part of the bud selection of vines and other fruiting plants which has been practiced in the past. It has been the common advice of books and specialists to keep "performance records" of individual plants of a variety and to use as a source of scions the individuals having the highest records for bearing.

A publication issued in 1906* states that: "A vineyard of pedigreed vines of all our most desirable varieties would be a most valuable acquisition for the State. Such a vineyard might be started with cuttings in the way described and each variety gradually brought up to its highest possible bearing capacity by grafting all the vines of each variety with cuttings taken from the vine of that variety which has shown the best and most regular bearing qualities during a term of years."

The Department of Agriculture of Victoria in 1924** gives the following advice:

"The improvement of the fruit-growing capacity of a variety by means of careful selection of cuttings is no new discovery; it has repeatedly been recommended by different officers of this Department, and its importance is now very generally recognized. It is a point, however, which was for many years much neglected by the majority of Victorian vine-growers, with the result that several of our vine varieties show more or less marked deterioration in their yield of fruit.

"In order to secure prolific scions, the best individual vines in a block of any given variety should be carefully marked—quality and quantity of fruit, as well as general health and vigor, are the essential points to be considered in the selection of these scion-bearing vines, which may best be carried out immediately before vintage. Only fruit-bearing canes on the vines thus selected should be used as scions."

This advice is evidently based on *a priori* reasoning from false analogy. The difference in egg production of two hens of the same breed is analogous to the difference in productiveness between two seedlings of a vine variety. In each case, the progeny individuals

* Bioletti, Frederic T. Selection and preparation of vine cuttings. California Agr. Exp. Sta. Circ. 26:4. 1906.

** Planting and reconstitution of vineyards. Unnumbered Circ. Dept. Agr. Melbourne, Victoria. Apr. 15, 1924.

are the result of a blend or assortment of the qualities of two parents (or with the plant of the reassortment of the qualities of a single heterozygous parent) and qualities of either parental factor may appear in the offspring. In a vine grown from a cutting, there are no parents in this sense. The new vine is not a new individual in the same sense as a chicken hatched from an egg. It is simply a part of a single individual—the clonal variety—which originated from a seed. All the millions of Muscat of Alexandria vines which are now growing in five continents are, from the point of view of heredity, simply parts of a single plant which originated from a seed, probably in Southwestern Asia many thousands of years ago.

The *a priori* assumption, therefore, should be that any bud taken from any vine of Muscat of Alexandria will produce a vine having all the possibilities of any and all vines of this variety. It follows from this assumption that a bud from a Muscat vine which has never borne a crop is just as likely to give us a heavy-bearing daughter vine as a bud from a Muscat vine which has a long record of large crops. The *onus probandi* lies with those who maintain the contrary.

The only demonstrated basis for the contrary conclusion is the possibility of bud mutations.

Occasionally there appears among Muscat vines, as among plants of other clonal varieties, an individual or unit showing some marked difference from the varietal complex of characters. Such a difference is, or may be, heritable in the sense that it is persistent and can be propagated by cuttings. Established cases of this kind are however rare in *Vitis vinifera* and only one well authenticated case has been noted in California among the 100,000,000 vines of the Muscat growing here. This is a case of "gigantism," similar cases of which have been observed in California with several other varieties of *Vitis vinifera*. The black and red variations of the Muscat of Alexandria may have originated as seedlings.

The great persistence of the characters of the Muscat is evident. What hope, therefore, is there that the bearing of Muscat vines can be improved by a careful selection of planting stock from mother vines having a high record for heavy bearing, and what danger is there from neglecting this selection?

INVESTIGATION

In the hope of obtaining an answer to these questions, an investigation was started at the Kearney experimental vineyard in 1911. A block of 1200 Muscat vines grown from unselected cuttings rooted

in a nursery in 1910 was planted in a piece of fairly uniform soil and a record of the crop of each vine kept for five years, 1914–18. In 1920, cuttings were taken from each of the 115 vines having an annual record of over 29 pounds (30 pounds–38 pounds) and of the 86 vines having an annual record under 16 pounds (7 pounds–15 pounds). These cuttings were rooted in the nursery and planted in a plot of very uniform soil in the Davis experimental vineyard in 1921.

The plot consists of 18 rows of 35 vines each. Every fourth row commencing with row 1 was planted with the progeny of low-bearing vines and the intermediate groups of three rows with the progeny of heavy-bearing vines. They were arranged so that the vines of lowest-bearing parentage adjoined the vines of highest-bearing parentage, i.e., vine 1 of rows 1, 5, etc., originated from a 7 pound parent and vine 1 of rows 2, 6, etc., from a 38 pound parent, and so on.

The four groups of four rows were made as nearly replicas as possible except that the largest and most vigorous rootings of both heavy and light bearing parentage were planted in the first rows, and the following rows planted with rootings in order of gradually diminishing size and vigor. There was, therefore, a continuous and gradual reduction in the size and vigor of the rootings from rows 1 and 2 to rows 17 and 18.

CORRELATION OF CROP AND PLANTING STOCK

This arrangement makes it possible to note any differences due to: (1) Bearing of the parent vines, and (2) quality of the rootings.

Effect of quality of rootings.—Before taking up the influence of the parent vines, the influence of the rootings will be considered.

There was a noticeable difference in the size and apparent quality of the rootings, but none of them were poor. Only two failed to grow and all the rest grew well. To a cursory glance, at the end of the second growing season, there was little difference of growth visible between the two ends of the plot.

The first crop worth harvesting was weighed in 1923, when the vines were in their third year. The result of the weighings by rows is shown in chart A.

Omitting rows 1 and 18, there is a fairly regular decrease of crop from one end of the plot to the other with no obvious influence ascribable to the parent vines. The influence of the size and quality of the rootings, on the other hand, is marked. The average crop of the vines in row 2, where the best rootings of heavy-bearing parentage were

grown, was 23 pounds, while that of the vines in row 16, where the poorest rootings of heavy-bearing parentage were grown, was only 11.7 pounds. A similar contrast is shown by the vines of low bearing parentage.

The best rootings (row 2) therefore yielded a crop of over 3 tons to the acre more than the fair rootings (row 16). The value of the extra crop was much greater than the total cost of the best rootings.

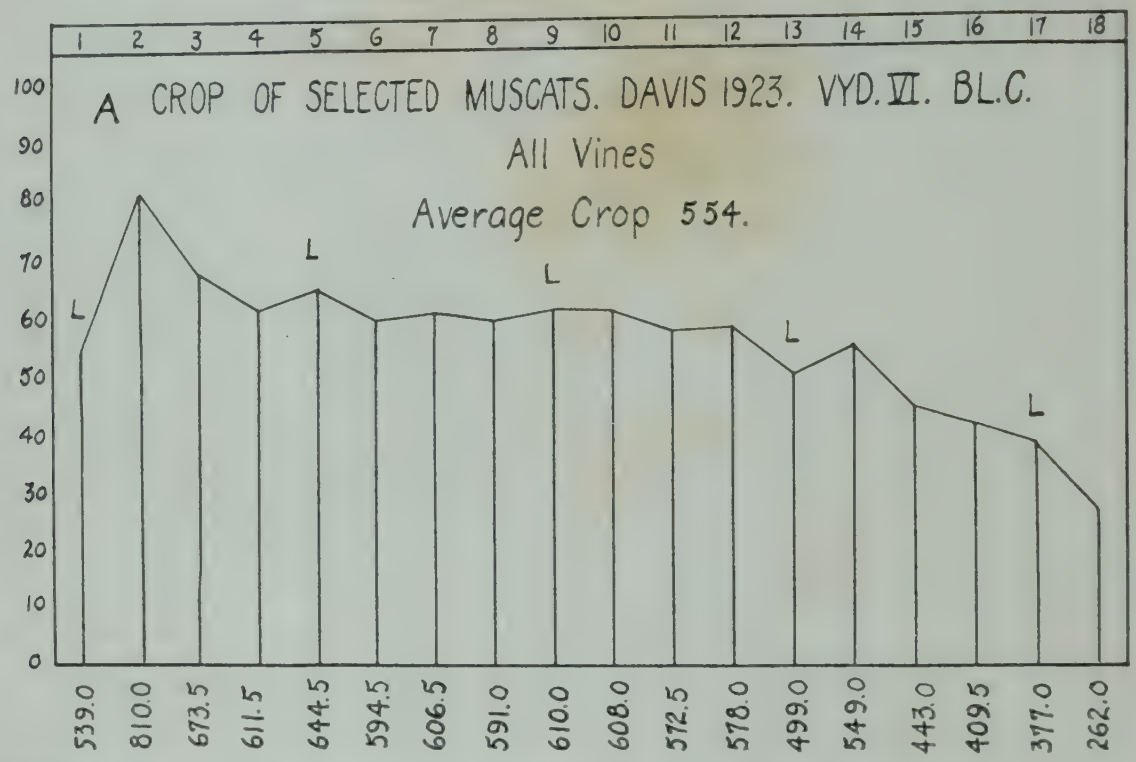

CHART A.—First (1923) crop of progeny vineyard. Rows of low-bearing parentage marked L.

All the vines looked strong and healthy so that it is probable that the difference is simply one of quicker development. Later crops have indicated strongly that the vines from the poorer rootings will finally produce as well as those from the best. The loss is merely one of time. The difference in development is shown by the average circumference of the vines in the spring of 1924 as appears in table I where it is compared with the corresponding difference in crop.

This table shows that the average circumference of the vines drops off from the north to the south of the plot. If we calculate the crop for each row in the ratio of the cube of the average circumference, we obtain a curve almost parallel with the actual crop curve except with the end rows 1 and 18. These rows are evidently abnormal in the amount of crop gathered, possibly on account of greater exposure

to wind during the blossoming season. There is plainly a direct correlation of both crop and size of vine with the size of the rooting at planting. This is made evident by the approach to parallelism of the crop and growth curves in chart B.

TABLE I

MEAN CIRCUMFERENCE OF TRUNK AND CROP OF VINES

Rows	Circumference, cm.		Crop in pounds per row	
			Actual	Calculated*
1**	L 12.64		539.0	636
2	12.82		810.0	664
3	12.79	−12.72	673.5	−698	659	−649
4	12.56		611.5	624
5	L 12.79		644.5	659
6	12.44		594.5	607
7	12.59	−12.23	606.5	−597	629	−581
8	11.67		591.0	508
9	L 12.32		610.0	589
10	12.26		608.0	581
11	12.15	−12.11	572.5	−586	565	−560
12	11.92		578.0	534
13	L 12.08		499.0	556
14	11.98		549.0	542
15	11.30	−11.64	443.0	−467	455	−518
16	12.09		409.5	557
17	L 11.61		377.0	493
18**	11.64		262.0	497
Means	12.21		573.5	570

* The figures in this column are what would have been obtained if the crop had been in proportion to the cube of the diameter.

** Rows 1 and 18 appear abnormal and are omitted in the calculation.

The influence of the quality of the rootings on the first crop is shown very clearly by chart B. The figures give the average crop of 35 vines for each of the 5 lots of vines with heavy bearing parents and of the 5 lots with light bearing parents. Each lot of the heavy bearers consisted of 3 rows except the 5th lot (row 18).

The lower solid line of the chart represents the crop of the vines of heavy-bearing parentage, the upper that of the vines of low-bearing parentage. They are very nearly parallel except at the upper end and to a less degree at the lower. The probable cause of these exceptions has already been discussed. The fact that the line of the vines of low-bearing parentage is higher than that of the others and drops less at the lower end is probably due to selection of rootings. Only one-third as many "poor bearers" as "heavy bearers" were planted and there was, therefore, a better selection of rootings. All the heavy bearers were planted, but there was a surplus of the poor bearers, which resulted in the rejection of the poorest rootings.

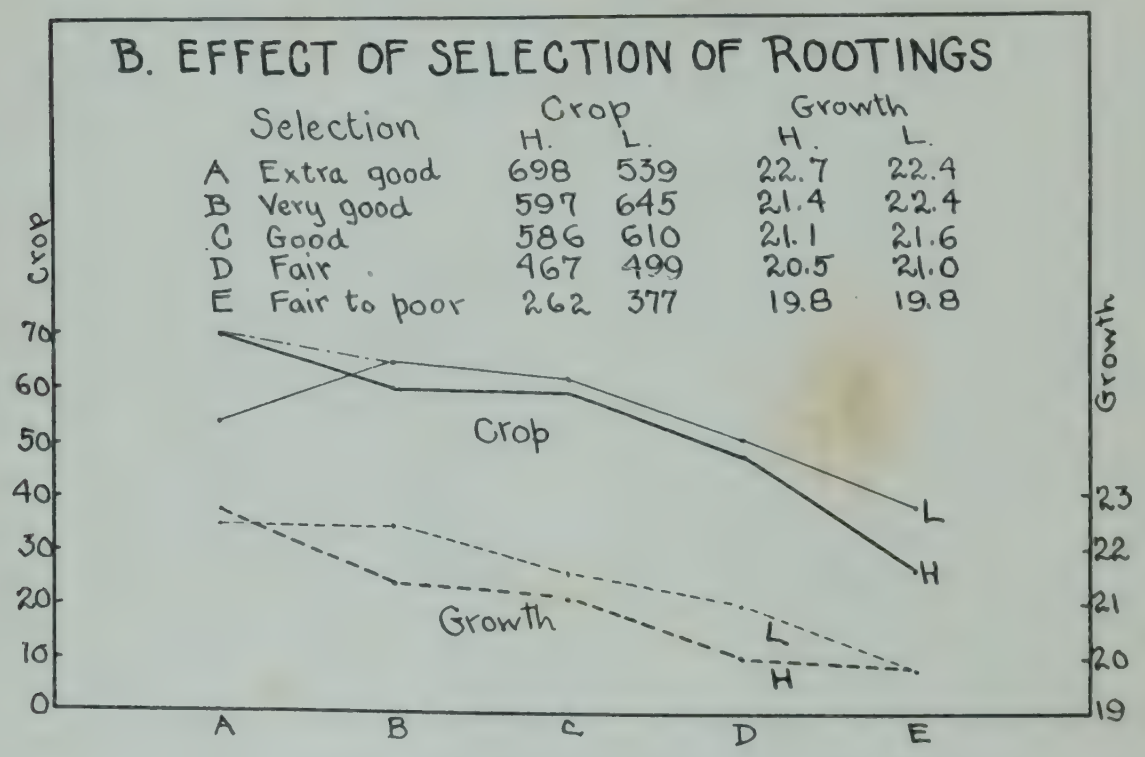

CHART B.—Effect of selection of rootings.

The dotted line at the upper end of the line for vines from poor bearers represents about the position that it would have taken if the first row had conformed with the general increase of bearing from south to north. It is assumed that the best rootings in the two cases were equally good.

Effect of the parent vines.—More than one crop is necessary to come to a definite conclusion regarding the influence of the parent vines on the bearing of the progeny, but it is interesting to find that there is no sign of any influence in the first crop. This can be seen clearly in chart C.

The solid line on the left shows the crops of vines of heavy-bearing parentage from row 2 to row 18. The average crops of the corresponding parent vines are shown by the dotted line below. A similar comparison is made for the vines of low-bearing parentage by the lines on the right. (The crops of the progeny vines are shown on a scale 3.5 times as large as that used for the parent vines.)

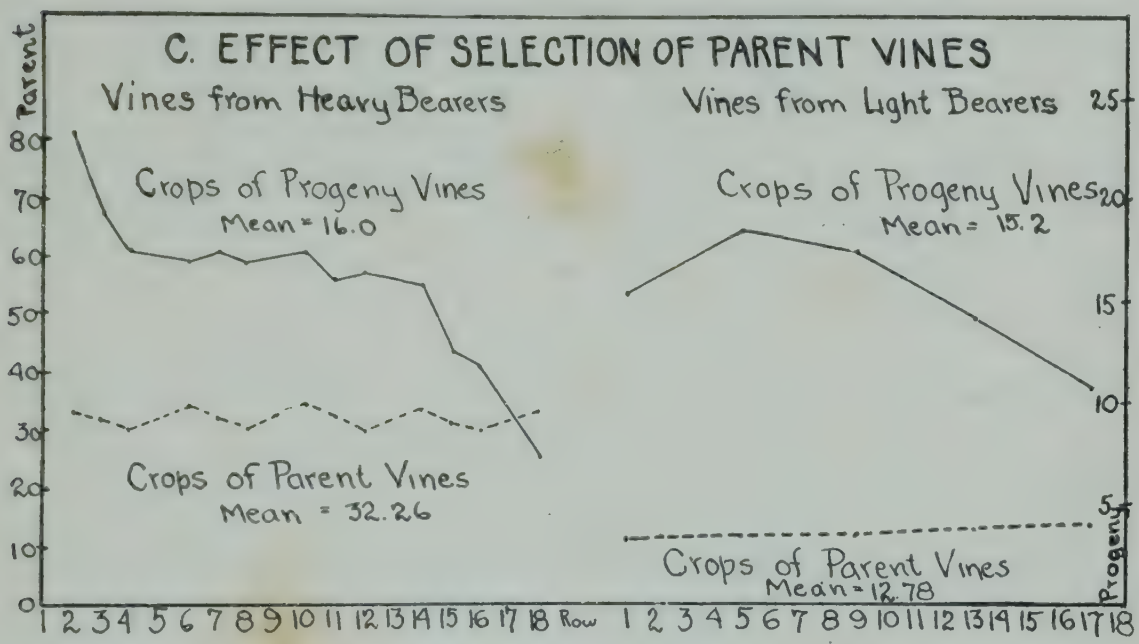

CHART C.—Effect of selection of parent vines.

There is an almost imperceptible falling off of crops of the heavy-bearing parent vines from north to south (left to right) and a more marked falling off from row to row of each lot of three, but there is no correspondence with the much greater falling off in the same direction shown by the crops of the progeny vines.

With the light-bearing parent vines, there is a slight increase of bearing from north to south coinciding with the marked decrease of bearing of the progeny vines.

RESULTS OF LATER CROPS

With the vintage of 1925, the progeny vineyard had yielded three crops. Table II shows the average crop in pounds per vine for each row, for each year, and for the three years.

The data of this table show evidence (1) of a general tendency to an annual alternation of higher and lower crops, (2) of a gradual recovery of the crops of the vines grown from the less vigorous rootings, and (3) a complete lack of correlation between the yields of the parent vines and those of the progeny.

Alternation of crops.—A distinct tendency to an alternation between heavier and lighter crops is shown by chart D(a). Row 18 which produced only 49 per cent of the average in 1923—the smallest crop of the year—produced 117 per cent in 1924—the largest for this year except row 8. This indicates that the smallness of the crop of row 18 in 1923 was not due to any defect of soil, but simply to the inferiority in size and vigor of the rootings planted, which delayed their development, and perhaps to some evanescent defect of position. With the second crop, this handicap had apparently been overcome. The small crop of 1923 allowed the vines to acquire the size and vigor necessary for the large crop of 1924. Similar and proportionate effects are shown by 15, 16 and 17, the other rows planted with the poorer rootings.

TABLE II

CROPS OF PROGENY VINEYARD, 1923, 1924 AND 1925. (MEAN CROP PER VINE FOR EACH ROW AND EACH YEAR.)

Row	1923	1924*			1925	Means of 1923–24	Means of 3 years
		a	c	b			
1 L	15.4	13.3	19.6	6.3	17.4	17.5	17.5
2	23.1	12.5	16.8	4.3	22.7	20.0	20.9
3	19.4	13.3	18.7	5.4	20.2	19.1	19.4
4	17.5	11.3	18.3	7.0	22.0	17.9	19.3
5 L	18.4	11.9	17.4	5.5	19.5	17.9	18.4
6	17.0	13.2	17.9	4.7	20.5	17.5	18.5
7	17.8	15.1	19.8	4.7	22.0	18.8	19.9
8	16.9	14.6	20.3	5.7	22.2	18.6	19.8
9 L	17.4	12.0	18.1	6.1	20.5	17.8	18.7
10	17.4	10.0	15.9	5.9	20.2	16.7	17.8
11	16.3	10.5	15.3	4.8	19.8	15.8	17.1
12	16.5	8.2	12.3	4.1	18.7	14.4	15.8
13 L	14.2	9.2	13.9	4.7	18.8	14.1	15.6
14	15.7	9.1	14.3	5.2	20.3	15.0	16.8
15	12.7	10.1	15.9	5.9	22.1	14.3	16.9
16	11.4	10.0	16.6	6.6	19.4	14.0	15.8
17 L	10.8	10.2	18.2	8.0	20.8	14.5	16.6
18	7.7	13.3	20.1	6.8	18.4	13.9	15.4
Means	15.9±.54 σ 3.41		17.2±.31 σ 1.92		20.3±.23 σ 1.42	16.5±.30 σ 1.94	17.8±.24 σ 1.51

a=1st crop, b=2nd crop, c=total=1st+2nd crops, for 1924.

* A spring frost in 1924 injured many of the first shoots. The first crop was consequently light. The loss was made up, at least in great part, by a second crop nearly half as large as the first. The second crop is borne on lateral shoots and main shoots which grow after the frost. It was negligible in 1923 and 1925 and was not weighed.

On the other hand, row 2 which produced 145 per cent of the average in 1923—the largest crop of the year—produced only 98 per cent in 1924.

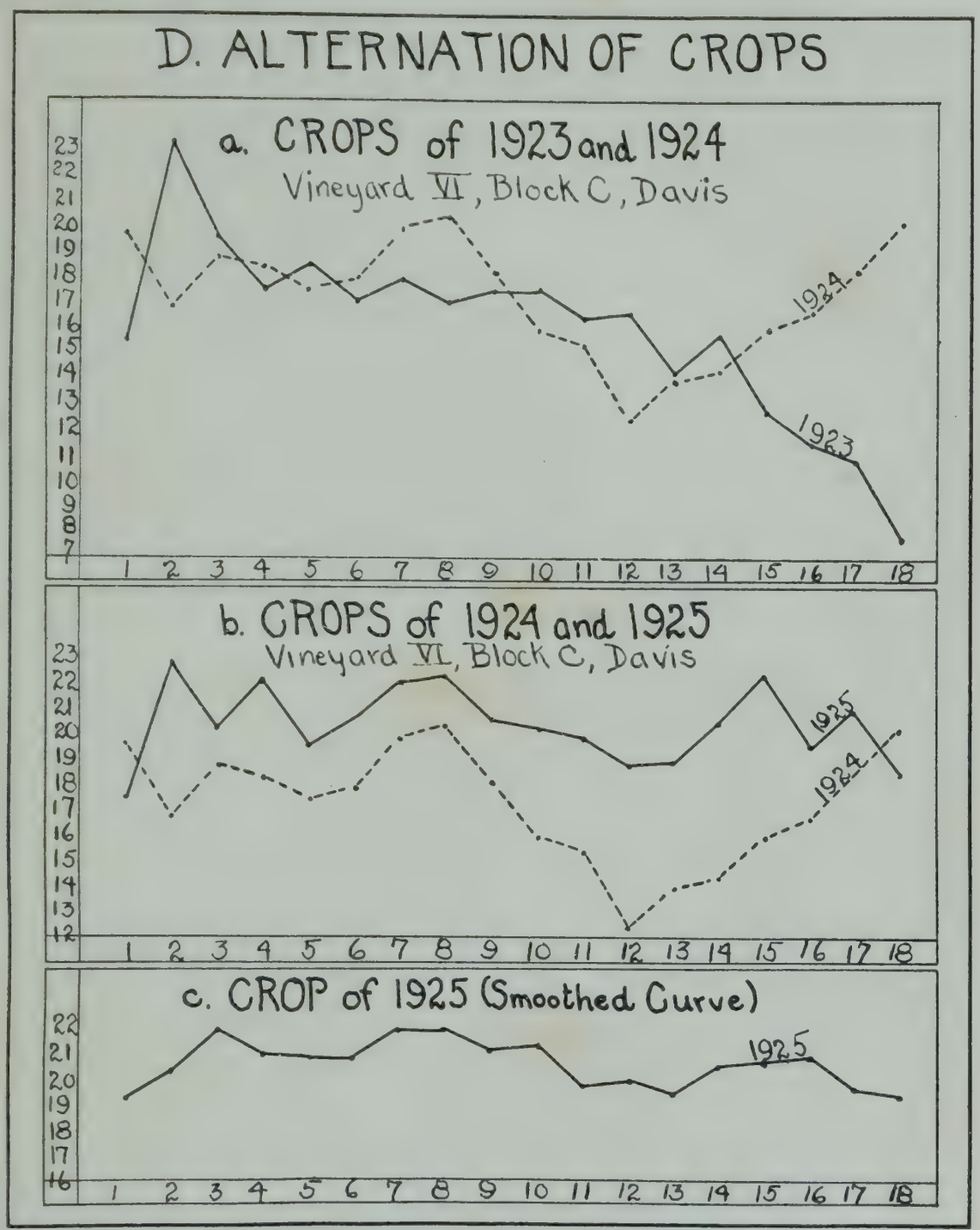

CHART D.—Alternation of Crops.

The case is not quite so clear with the intermediate rows, 3 to 14 inclusive, probably because the crops of these rows were nearer to the average, which may perhaps be considered the normal for the vines in these rows, where the alternation, due to over—or under—bearing, would be reduced to a minimum and thus be masked by the

effects of other factors which influence the crop. The graph indicates, however, that the 107 per cent of the average produced in 1923 by rows 6–9 planted with vigorous vines was less than their normal and, consequently, that they produced more than their normal or 111 per cent of the average in 1924. The reverse seems to have been the case with rows 10–14 planted with less vigorous vines which produced more than their normal or 101 per cent of the average in 1923, and, consequently, less than their normal or 83 per cent of the average in 1924.

Chart D(b) showing the crops of 1924 and 1925 indicates that the alternation of crops had to a great extent been overcome in all the rows with full development of the vines and by differential* pruning. The differences of crop shown by the different rows in 1925 are probably due chiefly to variable factors which affect the vines irregularly, such as frost, and defects of pruning and irrigation. The continued deficiency of crop in the neighborhood of row 12 may indicate some constant unfavorable factor such as a less suitable soil. A defect of exposure or other factor of position may account for the deficiency in average crop shown by row 1 as compared with rows 2, 3 and 4, and of row 18 as compared with rows 15, 16 and 17.

Ultimate effect of vigor of rootings.—It has been shown (see chart A) that there was a high correlation between the quality (vigor and size) of the stock rootings when planted and the first full crop, at the end of the third season of growth.

Table II shows that the deficiency in the yield of the vines grown from small rootings, marked in the first crop, tends to disappear in the second and third crops. This is indicated by the gradual decrease in magnitude of the standard deviation of 3.41 for the first crop to 1.92 for the second and to 1.42 for the third. (See table II.) This is shown graphically by chart D(c) which indicates little or no variation of crop at the third harvest ascribable to the original quality of the rootings.

Correlation of yield of parent with yield of progeny.—The second and third crops have confirmed the evidence of the first that the yields of the parent vines have no perceptible influence on the yields of the progeny vines. This appears clearly in table III in which the three crops of the progeny of low-yielding parents and of the progeny of high-yielding parents are segregated and compared.

* Differential pruning is the means used in pruning to increase the crop on vigorous vines and thus utilize their possibilities, and to decrease the crop on weaker vines and thus renew their vigor. It consists in pruning more or less severely inversely as the vigor of the vine. See: Some Common Errors in Vine Pruning and Their Remedies. California Agr. Exp. Sta. Circ. 248:4, 5. 1922.

The vines of high-yielding parentage produced in the three years an average annual crop of .5 pounds to the vine (3 per cent) more than the vines of low-yielding parentage. This difference is not significant because the coefficient of variability of all the vines was 1.51 ÷ 17.8 or 8.48 per cent. (See table 2.)

If we compare the crops of the vines derived from the highest-yielding parents with those derived from the lowest-yielding parents, we obtain a result of similar magnitude, but in the opposite direction.

This is shown in table 4 which compares the average crops of the progeny of the 37 and 38 pound parent vines with the progeny of the 7, 8, 10 and 11 pound parent vines. The difference in this case is .3 pounds or 1.5 per cent in favor of the vines of low-yielding parentage.

Mutations of productivity.—It is plain that mass selection of parent vines for productivity has had no perceptible influence on the yields of the progeny vines. No significant difference was found between the average yields of 540 vines derived from 30–38 pound parents and the average yields of 172 vines derived from 7–15 pound parents.

It remains to inquire whether among these high- and low-yielding parents one or more may represent a mutation of productivity. For this purpose, table 5 has been prepared showing (1) the performance of the progeny of the highest- and lowest-yielding parents, and (2) the parentage of the highest- and lowest-yielding progeny.

An examination of table 5 shows under A that the progeny of the 6 most fruitful parent vines having average crops of 37 or 38 pounds varied in average crops from 25 pounds to 5.8 pounds, and under B that the progeny of the 5 least fruitful parent vines having average crops of from 7 pounds to 10 pounds varied in average crops from 23.7 pounds to 10.1 pounds.

TABLE III

Crops of Progeny of Low-Yielding and of High-Yielding Parentage. Average Yield in Pounds per Vine

	1923	1924	1925	3-year mean
Rows 1, 5, 9, 13, 17 (low-yield parents)	15.2	17.4	19.4	17.4
All other rows (high-yield parents)	16.2	17.1	20.6	17.9

TABLE IV

CROPS OF PROGENY COMPARED WITH CROPS OF PARENTS

Parent vines		Progeny vines				
Mean crops for 5 years		Mean crops for 3 years				
Vines*	Crop	Vines*	1923	1924	1925	Mean
Heavy bearers:						
46:12	38	2:1	16	22	25	21
46:12	38	6:1	16	17	24	19
46:12	38	10:1	11	17	22	17
46:12	38	14:1	12	9	22	14
46:12	38	18:2	7	9	12	9
43:18	38	2:2	24	17	30	24
43:18	38	6:2	19	32	23	25
43:18	38	10:2	15	21	18	18
43:18	38	14:2	20	12	25	19
37:19	38	2:3	20	22	32	25
37:19	38	10:3	12	15	16	14
37:12	38	6:3	21	12	28	20
46:15	37	2:4	26	19	23	23
46:15	37	6:4	13	12	35	20
49:17	37	2:5	27	23	26	25
49:17	37	6:5	13	29	17	20
49:17	37	10:4	13	22	23	19
49:17	37	14:3	12	14	20	15
49:17	37	18:3	0	8	10	6
45:21	37	2:6	21	23	28	24
45:21	37	6:6	19	32	24	25
45:21	37	10:5	11	31	21	21
Means	37.6		15.8±.90	19.0±1.04	22.9±.85	19.3±.73
			σ 6.25	σ 7.25	σ 5.93	σ 5.05
Light bearers:						
22:23	7	1:1	9	11	20	13
22:23	7	5:1	21	10	22	18
22:23	7	9:1	6	6	19	10
22:23	7	13:1	10	10	33	18
16:17	8	1:2	18	27	27	24
16:17	8	5:2	16	19	16	17
21:22	8	1:3	15	30	17	21
21:22	8	5:3	20	27	25	24
21:22	8	9:2	6	15	32	18
16:15	8	1:4	19	17	25	20
29:18	10	1:5	12	27	21	19
33:1	11	5:5	21	16	32	23
33:1	11	13:2	16	19	30	22
33:1	11	1:9	23	20	21	21
50:7	11	5:4	15	21	14	17
41:23	11	1:8	26	26	32	28
Means	9.4		15.8±.98	18.8±1.19	24.1±1.18	19.6±.63
			σ 5.81	σ 7.08	σ 6.10	σ 3.70

* The numbers give the position of the vines in the experiment vineyards.

Table 5 shows further under C that the parents of the 9 most fruitful progeny vines having crops from 29 pounds to 25.3 pounds varied in average crops from 35 pounds to 13 pounds, and under D that the parents of the 9 least fruitful progeny vines having crops of from 1.3 pounds to 7.7 pounds varied in average crops from 33 to 13 pounds.

TABLE V

COMPARISON OF INDIVIDUAL PROGENY YIELDS WITH CORRESPONDING PARENT YIELDS

A. *Yield of progeny of the heaviest yielding parents*

Yield of parents	38	38	38	37	37	37	
Yield of progeny	20.8	23.4	24.3	22.4	25.0	24.1	
	19.1	25.0	13.7	19.3	19.3	24.7	
	16.3	17.7	20.0		19.1	17.4	
	14.2	18.7			15.1		
		9.2			5.8		
Means	17.6	18.8	19.3	21.4	16.9	22.1	Mean 18.9

B. *Yield of progeny of the lightest yielding parents*

Yield of parents	7	8	8	8	10	
Yield of progeny	13.3	23.7	20.3	20.2	18.7	
	17.1	16.7	23.5			
	10.1		17.3			
	17.4					
Means	14.5	20.2	20.4	20.2	18.7	Mean 18.0

C. *Yield of parents of the heaviest yielding progeny*

Yield of progeny	29.0	28.0	28.0	27.7	27.3	27.0	25.7	25.7	25.3	Mean 27.1
Yield of parents	14.0	13.0	33.0	33.0	30.0	31.0	33.0	31.0	35.0	Mean 28.1

D. *Yield of parents of the lightest yielding progeny*

Yield of progeny	1.3	1.7	3.0	6.0	6.6	7.0	7.3	7.7	7.7	Mean 5.36
Yield of parents	13.0	31.0	30.0	33.0	33.0	30.0	30.0	14.0	32.0	Mean 27.3

None of the 6 most fruitful parent vines gave rise to any of the 9 most fruitful progeny vines (see tables 5A and C) and none of the 5 least fruitful parent vines gave rise to any of the 9 least fruitful progeny vines (see tables 5B and D).

It might be contended that the easiest place to detect a mutation of high productivity would be in the part of the vineyard where the average crops were lowest, though this would be to give up the whole case of the "pedigree bud selectors."

Chart E shows the parent vineyard at Kearney. The part of the vineyard from about row 13 to row 24 has many missing vines, and many with records of low yields. Only four of the vines with high records are found in this area. These are shown on the chart by two crosses and two stars.

Table 6 shows the records of the progeny of the three of these vines from which progeny were obtained, and for comparison the records of the progeny of the three parent vines with low records growing nearest to the three high-yielding parents.

TABLE VI

CROPS OF PROGENY OF HEAVY-BEARING PARENTS FROM POOR AREA

Parents			Progeny		
Vine	Crop		Vine	Crop	Means
	Pounds			*Pounds*	
20.6	30		16:14	13.7	
20.6	30		16:15	16.4	
20.6	30		13:5	11.6	13.9
20:8	35		10:23	17.9	
20:8	35		6:23	22.4	
20:8	35		2:23	25.4	21.9
21:8	33		7:13	18.3	18.3
	Mean	32.7		Mean 18.0	

CROPS OF PROGENY OF LIGHT-BEARING NEIGHBORING VINES

Parents			Progeny		
Vine	Crop		Vine	Crop	Means
	Pounds			*Pounds*	
22:9	12		1:19	19.8	
22:9	12		5:15	15.7	
22:9	12		13:6	16.7	17.4
19:10	12		1:16	21.8	
19:10	12		5:12	20.2	
19:10	12		9:8	21.8	21.3
21:5	14		13:24	13.7	
21:5	14		17:12	15.7	
21:5	14		9:30	17.0	15.5
	Mean	12.7		Mean 18.0	

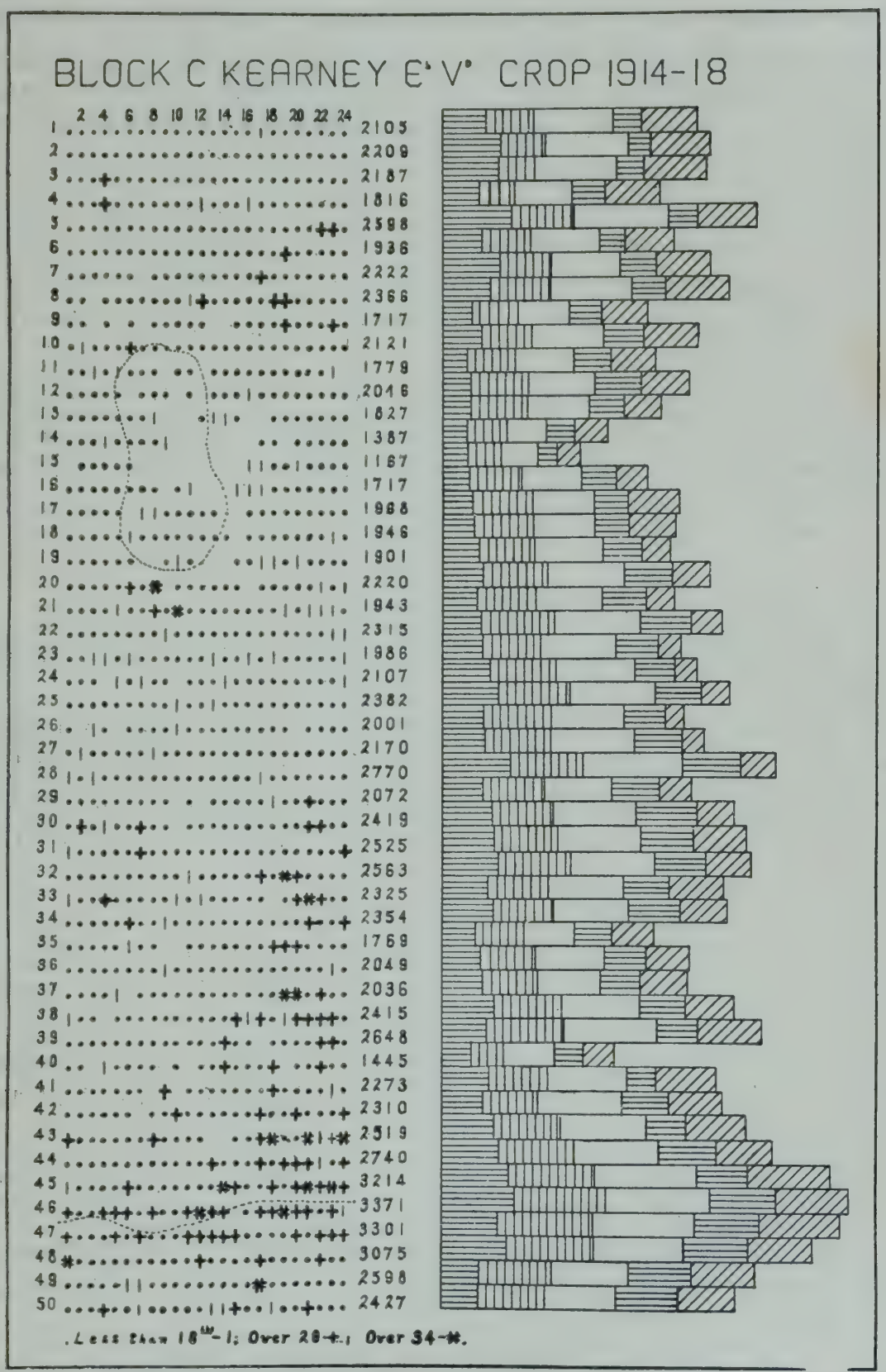

CHART E.—Parent vineyard at Kearney. The 5 crops are shown by variations of shading.

The high-yielding parents had records of 35, 33 and 30 pounds, the low-yielding parents 12, 12 and 14 pounds. The average yields of the progeny were 18 pounds in the case of the progeny of the high-yielding parents and the same—18 pounds—for the progeny of the low-yielding parents. The fact that the progeny of one of the high-yielding parents have the highest mean record—21.9 pounds—is offset by the fact that the progeny of another of the high-yielding parents have the lowest mean record—13.9 pounds. This record of 21.9 pounds is, moreover, considerably lower than the 28-pound record of the highest-yielding progeny which originated from an 11-pound parent. (See table 4.)

There is no evidence in this investigation, therefore, that the yield of the progeny vineyard would have been affected in the least weather the cuttings from which it was started had been taken from the heaviest-yielding vine in the parent vineyard or from the lightest.

SUMMARY

1. All commercial vineyards consist of clonal varieties propagated by buds.

2. It is the general belief of growers and of their advisers that a careful choice of "mother vines" on the basis of performance in the matters of yield, quality and other characters is necessary to maintain, and useful to improve, the fruitfulness and general utility of the variety.

3. It is generally recognized that the use as planting stock of a bud cutting or rooting which is well grown and well nourished gives the best results. Opinions differ as to whether vines from weak stocks which grow will finally equal vines grown from strong stocks.

4. To test the validity of these beliefs an investigation was started in 1910 at the Kearney experiment vineyard and continued at the Davis experiment vineyard until 1925.

5. An experiment plot of 1200 Muscat vines was planted at Kearney in 1911 with rootings grown from stock of unselected vines rooted in a nursery in 1910. This was called the "parent vineyard," and a record was kept of the yield of each vine during the 5 seasons of 1914–1918. (See chart E.)

6. In 1920, cuttings were taken from the 115 most productive vines in this parent vineyard and from the 86 least productive. The mean annual yield of the former was from 30 to 38 pounds and of the latter from 7 to 15 pounds. These cuttings were rooted in a nursery at Davis.

7. In 1921 an experiment plot of 627 vines was planted at Davis with the rootings obtained from the cuttings from the Kearney parent vineyard. This was called the "progeny vineyard."

8. Of the rootings of high-yielding parentage, 452 were planted in 4 groups of 3 rows each and 1 group of 1 row; and of the rootings of low-yielding parentage, 175 were planted in 5 groups of 1 row each, alternating with the groups of high-yielding parentage.

9. All rootings were carefully graded and arranged in planting according to quality, i.e., size and perfection of form. There was thus a continuous and gradual reduction in the quality of the rootings from one end of the plot to the other.

The arrangement was such that (*a*) the vines of highest-yielding parentage were contrasted directly with the vines of lowest-yielding parentage, and (*b*) the complication of vigorous vines growing near weak vines was avoided.

10. *Effect of quality of stock.*—

(*a*) The first crop of the vines from the strongest 25 per cent of the rootings was about 50 per cent larger than the first crop of the vines from the weakest 25 per cent. This difference was in great part reversed by the second crop and there was little difference in the third crop.

(*b*) The advantage of the strongest rootings was in reaching nearly full bearing the third season instead of the fourth as with the weaker rootings. The poorest rootings were all equal to what are usually considered No. 1 quality. With more imperfect rootings such as are very commonly planted, the difference would undoubtedly have been greater.

(*c*) The larger crop of the vines from vigorous rootings was accompanied by a larger diameter increase of the trunk.

11. *Effect of mass selection of parent vines.* No correlation was found between the crops of parent vines and the crops of progeny vines. Mass selection on the basis of yielding records had no perceptible effect. The average yields of vines derived from low-yielding parents were virtually equal to the average yields of vines derived from high-yielding parents and showed the same order of variability.

12. *Effect of individual selection of parent vines.* From the 6 highest-yielding parent vines were grown 22 progeny vines. Not one of the 9 highest-yielding progeny vines was among these 22.

From the 5 lowest-yielding parent vines were grown 11 progeny vines. Not one of the 9 lowest-yielding progeny vines was among these 11.

DISCUSSION AND CONCLUSIONS

The testimony of this investigation is that:

1. Exceptionally large and well formed vine rootings developed more quickly and produced a full crop one year sooner than ordinary good rootings.

2. The differences in bearing represented simply differences in rapidity of development and they almost disappeared with the third crop. With very inferior rootings they might never have disappeared, especially where rootings of various degrees of size and vigor were planted together. In such a case, the weak vines would be likely to be permanently inferior owing to competition with their more vigorous neighbors. This would bring about an irregularity of the vineyard which would probably persist and detract from its value. This is what occurs where missing vines are replaced in a vineyard even as early as the second year.

3 Mass selection of vine cuttings on the basis of the yields of the parent vines was of no value in improving or maintaining productivity.

4. The attempt to increase the bearing of a variety of vine by the selection of buds from a parent vine which has been distinguished by continuous and heavy bearing superior to that of the average or of any of the vines of the same variety but which shows no other distinguishing character is fruitless.

5. The attempts of nurserymen and others to preserve or to improve the productivity of clonal varieties of fruit trees by bud selection based exclusively on yield records of the parent plants would be wasted efforts if applied to vines. That it is of any use for this purpose for other fruit trees is doubtful.

OPPOSING EVIDENCE

The last two conclusions are opposed to the opinion of some investigators. Some hold directly contrary opinions, which, however, appear to be founded on a wrong interpretation of the evidence. Two of the most notable cases are those of Davis* who studied bud selection in apples, and those of Shamel who studied citrus fruits, especially oranges and lemons. Shamel** *et al.* in a recent account of an investi-

* Davis, M. B. The possibility of the transmission by asexual propagation of the high-yielding ability of individual apple trees. Scientific Agr. 2:120–124. 1921.

** Shamel, A. D., C. S. Pomery, and R. E. Caryl. Bud selection in the Washington Navel orange. Jour. Hered. 16:371–374. 1924.

gation of the results of bud selection of the Washington Navel orange states, "These results indicate that the quantity and quality of fruits produced by citrus trees are transmissible characters occurring as bud variations and as such are capable of perpetuation through budding." It is not quite clear what the authors mean by this statement. If their meaning is simply that the productivity of a clonal variety is transmissible to new plants propagated by buds, their statement is merely a truism. The whole practice of growing plants by vegetative multiplication is based on the well established belief that productivity in common with all other characters is a quality inherent in the bud and of the growth arising from this bud whether the growth is made in the form of a branch on the tree where it originated or in the form of another tree produced by budding, grafting or any method of propagation by vegetative segments.

If, on the other hand, they mean that all, or most, or many of the variations in yield of individual plants of a clonal variety are certain or even likely to appear in new plants propagated vegetatively from buds of these individuals, their opinion is ill-supported by the evidence they present.

If a Washington Navel orange gives rise to a branch of markedly different habit of growth with fruit of plainly different quality and yields inferior to those of the type, as in Shamel's experiment, it is an example simply of an ordinary and easily recognized "bud sport" or mutant. If these characters were not transmitted, it would be worthy of remark and require explanation. That they are transmitted does not render even probable that differences of yield unaccompanied by other appreciable differences would be transmitted in the same way. And yet this seems to be the only evidence submitted in support of the advice on which commercial bud selection associations have acted in keeping continuous and expensive records of individual trees for the purpose of improving the productivity of clonal varieties of orchard trees.

If the mutation is evident without yield records, they are unnecessary If the yield records are the only evidence of mutation, they are in the present state of our knowledge incompetent for the purpose. Yield records are not a means of detecting a mutation, but of testing the value of a mutation after it has been found by other means.

In Davis' experiments scions were taken from three Wealthy apple trees with mean annual yield records of 105, 79 and 41 gallons respectively. No other differences in the trees were noted except that the light-yielding tree was smaller and weaker than the others.

The scions were root grafted on Crab seedlings and planted in parallel rows—the row of low-yielding parentage between the other two rows. The results were:

1. Of the heavy bearers 24 per cent died, of the medium 18 per cent and of the light bearers 35 per cent.

2. The relative order of the progeny trees in regard to bearing was the same as that of the parent trees.

Yield records of the trees—

Actual differences:	Heaviest	Medium	Lightest
Parent trees—mean annual crop in gals.....	105	79	41
Progeny trees—mean annual crop in gals.....	57	48	35
Percentage differences:			
Parent trees—mean annual crop.....	100	75	39
Progeny trees—mean annual crop.....	100	84	61

Under the new conditions, the progeny of the heaviest-bearing tree have fallen off in yield 46 per cent, the progeny of the medium tree 39 per cent and the progeny of the lightest-bearing tree only 15 per cent. That the differences have not disappeared entirely may very plausibly be ascribed to the fact that the scions of the low-bearing tree were probably weak like the tree from which they were taken, as indicated by their greater death rate, and probably made less growth during the first two or three years than their more robust neighbors. The fact that the row of weak trees was planted between the rows of strong trees would tend to maintain their disadvantage in later years.

In the tests at Davis with Muscat, it required four years for fairly vigorous vines to overtake very vigorous vines growing under similar conditions. With apple trees which are slower in development and of less vigorous recuperative powers, it might require several years longer for a weak plant to overtake its stronger competitors. In the experiments described by Davis, it might never overtake them because of the difficulty of overcoming the handicap of a bad start in the proximity of its vigorous neighbors which would quickly take possession of more than their share of the soil and space available.

The evidence indicates that the higher yields of the progeny of the high-yielding parent trees was due to their higher initial vigor and that the difference tends to disappear. Davis' results are an argument in favor of vigorous propagation stock, but give little support to the theory that he was dealing with mutations of productivity.

These two cases of bud selection of fruit trees which are supposed to have resulted in the increased productivity of a clonal variety and which appear to have been conducted with an approach to the requirements of equal conditions and control checks are more probably to be explained in the one case as an ordinary bud mutation where, as usual, many characters varied and in the other as an example of delayed development, due to a start with ill-nourished stock and continued inferiority due to unfavorable conditions. In neither case is there any evidence of a bud mutation of productivity unaccompanied by any other difference or of the possibility of detecting such a mutation by crop records if it did exist.

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A STUDY OF RESISTANCE TO WESTERN YELLOW BLIGHT OF TOMATO VARIETIES*

JAMES W. LESLEY†

INTRODUCTION

Western yellow blight is an important disease of tomatoes prevalent in certain regions west of the Rocky Mountains. In California the loss from this disease is very heavy in certain years; for instance, in 1925 tomato growers in the interior valleys of central California lost from 75 to 95 per cent of their crop from this cause. In this paper, for brevity, "western yellow blight" is called "blight."

The practicability of controlling blight by the use of resistant varieties seems worthy of thorough consideration. The object of the present work is the discovery of varieties well adapted to the conditions where blight is severe or the development of such varieties by breeding. The present paper reports the reaction of certain varieties to blight, the results of three years' work on selection for blight resistance and some results of hybridization.

It was found that the varieties Dwarf Champion, Dwarf Aristocrat, Red Pear, and certain strains selected for blight resistance, are more resistant than the standard commercial varieties Stone and Santa Clara Canner. In a blight attack of moderate severity the resistant varieties and certain selected lines are about 25 per cent less susceptible than the standard varieties, but in attacks of extreme severity in early summer all of these have been nearly 100 per cent blighted. The dwarf character is closely associated with resistance to blight;

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† Assistant in genetics.

this indicates that blight resistance is in this case conferred by the gene responsible for the dwarf character or by a gene or genes closely linked with it.

In the warmer interior sections of California where this work was conducted, the plant affected with blight ceases to grow, and the midribs of the leaves and leaflets become twisted, sometimes through as much as 180°. Owing to a combination of rolling and folding of the lamina the under surface of the leaves tends to be exposed to view and their texture becomes stiff and leathery. The whole plant assumes a pale sulfurous color which may first be seen

Fig. 1.—Tomato plant of standard habit with typical symptoms of western yellow blight.

in the mesophyll at the base of the young leaves. The veins often become purplish but this symptom is especially variable. The affected plant ceases to flower, the fruit stops growing but usually becomes prematurely colored, and the seeds cease to develop. Examination of the root system shows decay, especially of the smaller roots, the cortical tissue being shriveled and inclined to slough off. A photograph of a blighted plant is seen in figure 1. Occasionally such plants recover by sending up healthy shoots from the leaf axils. In the field the diseased and healthy plants usually appear to be scattered almost at random and may even grow side by side in the same hill.

The cause of blight is not known and as yet it has not been possible to induce the disease artificially. In California, blight usually

makes its appearance after the first warm period, but new cases may appear in the field at any time during the warmer part of the growing season, that is from April to October. Late-planted fields are often less affected than early ones. The severity of the disease is subject to very wide seasonal variation. Thus at Riverside, California, the season of 1923 was one of relatively little blight, while 1924 was emphatically a "blight year." The disease is also subject to regional variation. For instance, it is much more severe in the southern San Joaquin Valley of California than in coastal sections. On the basis of a study of the disease in California and of the weather records in the tomato-growing sections west of the Rocky Mountains, Shapovalov[11, 12] found a close correlation between the amount of blight and the rate of evaporation of moisture. Low relative humidity, high temperature and considerable wind movement regularly accompany severe outbreaks of blight. This conclusion is in keeping with many of the known facts concerning the remarkable seasonal and regional variation in the severity of blight and is a substantial contribution to our knowledge of a disease which has baffled pathologists for twenty-eight years.

Shapovalov[11] found that shading the plants with muslin was the most effective means of control; by this means the amount of blight was reduced more than two-thirds.

Several studies of the reaction of varieties and selected lines to blight in the western United States have been reported. In Idaho, Henderson[2] tested 13 varieties and reported that all of them were about equally susceptible. According to Hungerford[4], strains selected in the eastern states for resistance to *Fusarium* wilt were less resistant to blight in Idaho than local varieties, but certain other varieties and some selections from John Baer and Earliana showed marked indications of resistance. In Oregon, McKay[8] records that four varieties, including Norton, were all susceptible. In the State of Washington trials were initiated in 1903 and Humphrey[3] reports that certain varieties, Livingston's Dwarf Champion for example, were less susceptible to blight than others. Yaw[14] states that Dwarf Champion gives some indication of resistance when grown in California. Humphrey's and Hungerford's data, although not quantitative, suggest the feasibility of the control of blight by the use of resistant varieties.

ACKNOWLEDGMENTS

It is with pleasure that the author tenders his thanks to Mr. Michael Shapovalov, Pathologist, Office of Cotton, Truck and Forage-crop Disease Investigations, U. S. Dept. Agric., for much help in this work. Acknowledgments are also gladly made to Professor E. B. Babcock, Division of Genetics, University of California, who initiated this work, to Dr. J. T. Barrett, Associate Director of the Citrus Experiment Station, for subsequent help and to Mr. W. B. Camp, Associate Agronomist, U. S. Dept. Agric., for facilities provided at Shafter.

METHODS

The present work which was begun in the summer of 1922 was conducted in California. It was decided to test first the reaction to blight of numerous varieties and the efficacy of selection for blight resistance, and subsequently if necessary to employ hybridization with a view to increasing resistance.

In testing resistance a serious difficulty had to be faced at the outset, namely, our inability to induce blight artificially and our dependence on the irregular and highly variable severity with which it appears in different years. This difficulty adds considerably to the labor and time involved in working with blight.

In 1922 variety trials were visited at Zelzah†, Manteca, San Jose and Santa Ana, but so little blight appeared at any of these places that no comparisons were possible and it could only be concluded that certain of the varieties grown were not entirely immune to blight. In other places large commercial plantings of three of the most widely grown varieties, namely, Stone, Santa Clara Canner* and Earliana were found to be, in many cases, 75 per cent blighted.

In all the trials conducted by the writer the seeds were sown in cold frames and the plants transplanted to the field and grown with irrigation at a spacing of about 6 by 6 feet. The vines were not pruned and no manure or fertilizer was applied. The amount of blight on the plots was usually recorded at intervals of from 3 to 4 weeks. The unit plot was one row of varying length extending in the same direction as the irrigation furrows. Some of the varieties

† The trial at Zelzah was conducted by the Division of Genetics, at Manteca by the Division of Plant Pathology, University of California; at San Jose by the California Packing Corporation; at Santa Ana by the Haven Seed Company.

* This name has been given to the variety sometimes known as "Jap Canner" by Mr. Frank Dixon with the authority of the Canners League of California.

and selected lines were planted in plots replicated 2 to 4 times and in tables 2, 3 and 4 the result of each plot is shown separately. The proportion blighted was obtained by dividing the number of plants blighted by the number of plants which survived transplanting and were definitely classified. Stone or Santa Clara Canner was planted in the check plots; these varieties are very susceptible to blight and to about the same degree. Varieties and selected lines which showed more resistance than the check varieties are termed resistant.

That large differences in the proportion blighted may arise from causes which are not genetic was evident from a comparison of replicate rows of the same variety.

Near San Jose, one field of 20 acres of the variety Santa Clara Canner was for the greater part about 10 per cent blighted, but over an area of 4 to 5 acres as much as 80 per cent was blighted. The healthy plants were, as usual, scattered among the blighted ones.

It was clearly necessary to know something of the distribution of blight in fields containing only one variety.

Blight counts were made in two fields of the variety Stone. The variant was the proportion blighted in a single row of a certain fixed length, the rows running parallel to the irrigation furrows as in the variety trials. The proportion rather than the number blighted was taken because a variable number of plants were missing. If the proportion blighted per row were subject only to the fluctuations of simple or random sampling, the distribution would be of the binomial type of which the standard deviation is $\sqrt{\frac{p\,q}{n}}$ where p and q are the proportion blighted and not blighted, respectively, and n is the number of plants.

In the field near Arlington, California, a blight count was taken on November 20, 1924, on a block of 40 rows. The mean number of plants per row was 46, and the mean proportion blighted was 17 per cent. The standard deviation of the proportion blighted was 5.4 per cent; with one exception all of the sample rows came within a range of four times the standard deviation and the frequency curve was unimodal and approximately symmetrical. The standard deviation of random sampling in a binomial series where $n = 46$, $p = 17$ per cent and $q = 83$ per cent, is 5.5 per cent, or practically the same as that derived from the field data.

The other field was at Norco, California, and blight counts were taken on August 6 and again on September 30, 1925. A mere glance at this field showed that the blighted plants were much less evenly distributed than in the Arlington field. The area counted contained

32 rows; the average number of plants per row was 44 or approximately the same as at Arlington. The mean proportion blighted was 57 per cent, the attack being much more severe than at Arlington. The standard deviation was 10.1 per cent while that of random sampling was only 7.5 per cent; the difference is significant being 2.8 times the standard error of the standard deviation of random sampling. Furthermore there was a progressive change in the proportion blighted from one row to the next. These data indicate that the distribution of blight differs in different fields, that the variance in some cases may be similar to that of random sampling or in other cases may exceed it; also that the proportion blighted may change systematically from one row to the next, resulting in an unequal distribution over the field. Thus in interpreting observed differences in the proportion blighted in any given trial the ordinary test of significance may be inadequate, especially where the number of plants is small and the plots are not replicated.

In the statistical treatment of the data, each variety is compared with another variety, usually the check variety, and the probability that the difference in the proportion blighted might arise as a fluctuation of simple sampling was determined by Pearson's χ^2 method, the usefulness of which has recently been emphasized by Fisher[1]. This was equivalent to the application of this method to test the independence of variety and condition with relation to blight. Thus the number of blighted and not blighted plants expected in each of the two varieties to be compared was first ascertained assuming independence. Since the difference between the observed and the independence value was the same in each there is one degree of freedom. The value of P was taken from tables given by Yule[15] (p. 386) and in Pearson's Tables[13] (p. 30, Table XV C). The value of P then is the probability that a difference in the proportion blighted as great or greater than that observed might arise on random sampling assuming independence of variety and condition as to disease. As a rule, for a single experiment, a value of P exceeding .0027 ($\chi^2 = 9$) was not considered significant, and doubtless this rule excludes some significant differences. In tables 2, 4 and 6 the value of P attached to a given variety, unless having superscript,* refers to a difference in the direction of less susceptibility than the check variety.

In those cases where repeated trials clearly indicated resistance an attempt was made to measure its degree by another method, since the χ^2 method tests only the significance of an association and not its amount. For this purpose the standard error of the difference com-

puted from the binomial formula was compared with the observed difference in the proportion blighted. Thus if n_1, n_2 are the numbers of plants in the two varieties compared, p_0 is the proportion blighted and q_0 the proportion not blighted in the two varieties together, the standard error of the proportions in each variety is given by:

$$e_1^2 = \frac{p_0\ q_0}{n_1} \qquad e_2^2 = \frac{p_0\ q_0}{n_2}$$

and that of the difference is

$$e_{12}^2 = p_0\ q_0\ \left(\frac{1}{n_1} + \frac{1}{n_2}\right)$$

Assuming a normal distribution and that the direction of the difference was known, the probability of the observed difference arising as a fluctuation of simple sampling was taken from Pearson's Tables.[13]

TRIALS WITH VARIETIES AND SELECTED LINES

Experience in the year 1922 had emphasized the importance of choosing for trials localities where blight was as regular and severe as possible in its occurrence. With this in view the plots were planted in 1923 at the Citrus Experiment Station, Riverside, and at Shafter near Bakersfield. In 1922 a field of the variety Stone near Riverside and one near Bakersfield were respectively 30 per cent and 75 per cent blighted. Reports of previous years also indicated that as a rule the losses from blight at the latter place were especially severe.

Since the object of the trials in 1923 was the detection of resistance rather than its measurement a comparatively large number of varieties and progenies of single plants were included. Practically the same set of varieties and selected lines were set out at Riverside and Shafter. Stone was planted as the check variety.

At Riverside 33 varieties and 23 progenies of single plants selected in 1922 at Bakersfield, Fresno and San Jose were set out and an almost perfect stand was obtained. Out of 2475 plants recorded only 31 or 1.3 per cent blighted, so that this trial gave no comparative data and merely served to show that many of the varieties were not immune.

At Shafter the transplanting was done on May 11 and 12 in two separate fields (fields A and B in Table 1). Unfortunately many plants failed to become established and on June 6 much replanting was done in both fields. The blight attack was moderately severe but varied widely in plantings made in different fields or on different dates. The numbers are too small to permit the use of the χ^2 test and only permit conclusions of a most tentative kind.

TABLE 1

WESTERN YELLOW BLIGHT OF TOMATOES AT SHAFTER, CALIFORNIA, IN 1923
(Whole Season)

	Place	Date of trans-planting	Total number of plants recorded	Total number blighted	Per cent blighted
Stone	Shafter Field A	May 12	27	14	52
Selection 52	Shafter Field A	May 12	40	19	48
Selection 58	Shafter Field A	May 12	18	10	56
Selection 60	Shafter Field A	May 12	35	21	60
Selection 66	Shafter Field A	May 12	29	15	52
Selection 73	Shafter Field A	May 12	12	4	33
Selection 77	Shafter Field A	May 12	21	14	67
Selection 78	Shafter Field A	May 12	10	7	70
Selection 81	Shafter Field A	May 12	26	18	69
Selection 82	Shafter Field A	May 12	21	12	57
Stone	Shafter Field A	June 6	21	3	14
Dwarf Champion	Shafter Field A	June 6	39	3	8
Globe	Shafter Field A	June 6	10	3	30
Stone	Shafter Field B	May 11	23	1	4
du P. L. M.	Shafter Field B	May 11	14	3	21
Dwarf Aristocrat	Shafter Field B	May 11	21	0	0
King Humbert	Shafter Field B	May 11	16	7	44
Matchless	Shafter Field B	May 11	13	1	8
Magnus	Shafter Field B	May 11	10	3	30
Norduke	Shafter Field B	May 11	31	6	19
Perfection	Shafter Field B	May 11	16	3	19
Red Pear	Shafter Field B	May 11	10	0	0
Red Plum	Shafter Field B	May 11	26	3	12
Yellow Cherry	Shafter Field B	May 11	10	1	10
Yellow Peach	Shafter Field B	May 11	16	5	31
Yellow Plum	Shafter Field B	May 11	28	6	21
No. 213 (from Mexico)	Shafter Field B	May 11	15	2	13
Selection 53	Shafter Field B	May 11	21	0	0
Selection 55	Shafter Field B	May 11	10	0	0
Selection 75	Shafter Field B	May 11	15	2	13
Idaho Selection 3/2–1	Shafter Field B	May 11	17	3	18
Idaho Selection 3/2–2	Shafter Field B	May 11	13	0	0
Stone	Shafter Field B	June 6	66	2	3
Burwood	Shafter Field B	June 6	40	1	2
Dwarf Stone	Shafter Field B	June 6	46	3	7
Globe	Shafter Field B	June 6	48	2	4
Golden Dwarf Champion	Shafter Field B	June 6	33	2	6
Matchless	Shafter Field B	June 6	32	0	0
Norduke	Shafter Field B	June 6	40	1	2
Norton	Shafter Field B	June 6	36	5	14
Morse's San Jose Canner	Shafter Field B	June 6	39	7	18

In field A none of the selections were outstanding in blight resistance. Selection 66, the progeny of a healthy plant which had grown in the same hill, intertwined with a blighted plant, was as much blighted as the check. The smallest proportion blighted was in selection 73, from Santa Clara Canner. Further selections were made in this strain and seed was saved for the next year's trial.

In the second planting in field A Dwarf Champion was somewhat less affected than Stone.

In field B (Table 1) blight was much less prevalent than in field A, only four miles distant, and its distribution over the field was more uneven, possibly owing to soil differences resulting from the recent grading of the land. While most of the varieties and selected lines in the first planting (May 11) were affected by blight, Dwarf Aristocrat, Red Pear and line 53 from Stone and a few others were free from it.

In 1924 duplicate trials were again planted at Riverside and Shafter, including the more promising varieties, some progenies of single plants in the more promising selected lines of the previous year, and a few F_1 and F_2 hybrid progenies from crosses between standard and dwarf varieties. Excellent stands were obtained and in these trials over 3000 plants came under observation. Stone and Santa Clara Canner were used as check varieties. At Riverside the variety, Stone, which in 1923 was only 1.3 per cent blighted during the whole season, on July 3, 1924, was 21 per cent blighted. The disease continued to increase, on the whole with diminishing rapidity, until the middle of October. In the final count Stone was 50 per cent and Santa Clara Canner 55 per cent blighted. None of the varieties or selected lines were immune but notable differences were seen in the proportion blighted in different varieties and selected lines.

Table 2 shows the results at Riverside when each variety is compared with the check variety Stone as to numbers of healthy and blighted plants at the close of the first part of the season, up to July 3 inclusive, and as to the corresponding numbers for the whole season.

On July 3, in accordance with the indications at Shafter in the preceding year Dwarf Champion and Dwarf Aristocrat, the former especially, seemed to be resistant. When the figures for all three dwarf varieties, Dwarf Champion, Dwarf Aristocrat and Dwarf Giant, are compared with the check variety the resistance of dwarf varieties is emphasized. Red Pear, of standard habit, while again less affected than the checks, gave only a vague indication of resistance. Selected line 73–1, also of standard habit and derived from Santa Clara Canner, gave an indication of resistance in accordance with the record of 1923 (Table 1).

At the end of the season, corresponding with the increase of blight, the differences among varieties and strains were greater than on July 3. Indications of resistance seen earlier in the season were confirmed. Dwarf Champion again showed the most significant difference when compared with the check variety, and Dwarf Aristocrat continued to give an indication of resistance. Selection 73–1 still appeared to be resistant and 73–2 gave some such indication, but it

should be noted that the data in both cases are derived from single plots. Red Pear now gave more definite evidence of resistance than in the first period, whereas a single plot of Yellow Cherry which reacted much like Stone in the first period now gave evidence of even

TABLE 2

BLIGHT AT RIVERSIDE, CALIFORNIA, IN 1924

	Whole season							July 3			
	Per row		Total					Total			
	Number recorded	Per cent blighted	Number recorded	Number blighted	Per cent blighted	Compared with Stone	Probability that the difference from Stone is *not* significant.	Number blighted	Per cent blighted	Compared with Stone	
						χ^2	*P*			χ^2	*P*
Stone	56	55									
	43	53									
	36	39	135	68	50			28	21		
Burwood			53	21	40	2	.16	8	15	1	.32
Dwarf Aristocrat	53	34									
	42	40	95	35	37	5	.02	10	11	5	.02
Dwarf Champion	53	23									
	28	39									
	42	29	123	35	29	13	.0003	10	8	8	.005
Dwarf Giant			18	6	33			1	6	2	.16
Dwarfs (3 combined)			236	76	32	13	.0003	21	9	10	.002
Globe			25	9	36	2	.16	2	8	3	.08
Manx Marvel	52	65	78					20	26	0	1.00
	26	42		45	58	1	.32*				
Matchless			47	27	58	0	1.00	16	34	4	.04*
Norton			53	28	53	0	1.00	13	25	0	1.00
Red Pear	29	24									
	45	38	74	24	33	7	.01	10	14	1	.32
Santa Clara Canner	48	54									
	44	55	92	50	55	0	1.00	25	27	2	.16*
Yellow Cherry			56	36	65	4	.04*	10	18	0	1.00
Selection 52–1			44	19	43	0	1.00	8	18	0	1.00
Selection 52–2			47	22	47	0	1.00	11	23	0	1.00
Selection 52–3a			49	25	51	0	1.00	9	18	0	1.00
Selection 73–1			52	14	28	9	.003	7	13	2	.16†
Selection 73–2			51	18	35	4	.04	8	16	1	.32
Selection 267			36	13	36	2	.16	7	19	0	1.00

† Compared with Santa Clara Canner July 3 $\chi^2=4$, $P=.04$; whole season, $\chi^2=10$, $P=.002$.
* In the direction of *greater* susceptibility.

greater susceptibility. Norton, a selection from Stone resistant to *Fusarium* wilt, showed no greater resistance to blight than the parent variety. On the whole the data of Table 2 confirm the observations of the previous year. They also indicate in most cases a similarity in the reaction of a variety in these two overlapping periods.

At Shafter on May 7, 1924, 1200 plants were transplanted and of these 92 per cent became established. The incidence of blight was remarkable for earliness and severity. As early as June 3, 67 per cent of the check variety Stone had blighted. The younger leaves of affected plants in some cases showed wilting, presumably due to the extraordinarily dry, hot weather associated with this attack. In Table 3 the number of plants healthy and blighted on June 3 and on July 31 are compared with the corresponding number in the check variety Stone. As in previous trials, the data indicate that Dwarf Champion and Dwarf Aristocrat are more resistant than Stone. On the contrary, except for 73–1, the data for the selected lines conflict with the data of Table 2. As at Riverside, data derived from single small plots are of course inconclusive but the high proportion blighted in 73–2 in Table 3 indicates that this line is not resistant. If the onset of blight had been checked on June 3 (and according to Shapovalov,[11] such a check might be expected as a result of a suitable change in the climatic conditions), the difference between varieties observed on that date would probably have persisted throughout the season. For, as previously noted, the data at Riverside (Table 2) indicate that as a rule a positive correlation may be expected between the proportions blighted in the earlier and in the later portions of the season.

On June 30, out of 1107 plants recorded in all, 1091, or 98.6 per cent, were blighted and the differences so apparent at the earlier date were then negligible. Most of the survivors were dwarfs. A month later all but five of the plants were blighted. These survived throughout the season but one of them for some reason produced little fruit and scarcely any viable seed. It was evident that no variety or selection included in this trial could withstand an attack of such severity as occurred in this test. Dwarf Champion, which had shown resistance on June 3 and which has been reported resistant by Humphrey in the State of Washington was 99 per cent blighted on July 31.

In 1925 trials were again planted practically in duplicate at Riverside and Shafter. Many of the selected lines were derived from selections for blight resistance made in 1922, reselected in 1923 and 1924. Santa Clara Canner was used as a check. Field observations had indicated that this variety was about as susceptible as Stone and the data at Riverside (in Tables 2 and 4) confirmed this.

The trial at Riverside was planted in the same field as in 1924 but covered a larger area. Unfortunately, a comparison of replicate plots showed that those on the new land taken in developed con-

siderably more blight than corresponding plots on the old ground, although the whole trial was planted on the same day and received similar treatment. As a rule the plots were repeated serially in the same order: this proved to be by no means an ideal disposition but certainly tended to correct the error arising from the marked

TABLE 3

BLIGHT AT SHAFTER, CALIFORNIA, IN 1924

	June 3							July 31	
	Per row		Total					Total	
	Number recorded	Per cent blighted	Number recorded	Number blighted	Per cent blighted	Compared with Stone	Probability that the difference from Stone is *not* significant.	Number blighted	Per cent blighted
						x^2	P		
Stone	46	63							
	50	72							
	49	63							
	50	68	195	130	67			195	100
Burwood			49	31	63	0	1.00	49	100
Dwarf Aristocrat	41	61							
	41	27	82	36	44	12	.0005	81	99
Dwarf Champion	43	51							
	49	33							
	46	41							
	47	36	185	74	40	26	4×10^{-7}	183	99
Dwarf Giant			15	6	40			15	100
Dwarfs (3 combined)			282	116	41	29	10^{-7}	279	99
Manx Marvel			49	31	63	0	1.00	49	100
Norton			49	30	61	0	1.00	49	100
Red Pear			48	23	48	5	.02	48	100
Yellow Cherry			47	16	34	16	.00006	47	100
Selection 52–1			43	19	44	8	.005	42	98
Selection 52–2			27	7	26	18	.00002	27	100
Selection 52–3a			48	18	37	13	.003	48	100
Selection 73–1			44	14	32	20	8×10^{-6}	44	100
Selection 73–2			40	26	65	0	1.00	40	100
Selection 267			45	17	38	14	.0002	44	98

inequality in the distribution of blight. In Table 4, the upper part shows the results at Riverside and Shafter for the whole season of 1925.

Previous evidence of resistance in Red Pear is greatly strengthened by this trial. Line 310, the progeny of a first-year selection from Dwarf Champion, seems resistant, thus tending to confirm previous experience with this variety. All the four selected lines (73–1–2,

73–1–4, 73–1–5, 73–1–6) derived from 73–1, which gave evidence of resistance in both of the 1924 trials, gave evidence of resistance in 1925 also. Line 52–1–1, which was the progeny of the only standard plant in the whole trial which survived and set seed at Shafter in 1924, also gave an indication of resistance. Two short rows (only 49 plants) of the so-called Red Currant tomato (*L. pimpinellifolium*) were

TABLE 4

BLIGHT AT RIVERSIDE, SHAFTER AND DAVIS IN 1925

	Place and period	Per row		Total					Probability that the difference from the check variety is *not* significant.
		Number recorded	Per cent blighted	Number recorded	Number blighted	Per cent blighted	Check variety	Compared with check variety	
								χ^2	P
Santa Clara Canner	Riverside,	46	35						
	whole season	39	36				Santa		
		31	48				Clara		
		22	50	138	56	41	Canner		
Dwarf Champion,	"	50	22						
Selection 310		44	18						
		37	22	131	27	21	"	12	.0005
Norton	"			25	10	40	"	0	1.00
Red Currant (L. pimpi-	"	28	79						
nelli folium)		21	67	49	36	73	"	16	.00006*
Red Pear	"	50	12						
		43	14						
		37	24	130	21	16	"	19	.00001
Stone	"			61	32	52	"	2	.16*
Selection 52–1–1	"	48	21						
		41	28	89	22	25	"	7	.01
Selection 73–1–2	"	45	18						
		39	38						
		27	45	111	35	32	"	2	.16
Selection 73–1–4	"	44	23						
		39	41	83	26	31	"	2	.16
Selection 73–1–5	"	46	24						
		40	32	86	24	28	"	4	.04
Selection 73–1–6	"	43	14						
		42	33	85	20	24	"	7	.01
Santa Clara Canner	Shafter, to June	72	97						
	22	75	95	147	141	96			
Dwarf Champion,	"	73	78						
Selection 310		72	89	145	121	83	"	12	.0005
Red Pear				73	65	89		3	.08
Stone	Davis,								
	whole season			104	86	83			
Earliana	"			915	828	91	Stone	6	.01*
Globe	"			75	53	71	Stone	14	.0002
	"						Earliana	72	Less than 10^{-7}

* In the direction of greater susceptibility.

planted; one of these came from seed kindly sent by Dr. Weberbauer* and collected by him in Peru. This small fruited wild tomato seems to be more susceptible to blight than the cultivated *esculentum* variety used as a check.

As a result of three seasons' trials, the resistance of Dwarf Champion, Dwarf Aristocrat, Red Pear and selected lines 73–1–2, 73–1–4, 73–1–5 and 73–1–6 is considered to be well established.

Table 4 includes the records of three varieties grown at Davis, California, in 1925.† Blight was exceedingly prevalent. Earliana showed greater susceptibility than Stone or Globe. The latter appeared to be somewhat less susceptible than Stone, but further trial is needed.

In the Shafter trials of 1925 the transplanting was done on April 28 and a little replanting on May 4. About two weeks later a series of dust storms swept the field. On May 28 there was undoubtedly some blight but the little plants were so coated with dust that classification was difficult. On June 22 out of 1300 plants transplanted, only 63 were not blighted. As Table 4 shows, the check variety Santa Clara Canner was 96 per cent blighted, and again Dwarf Champion and Red Pear gave some indication of resistance. Selection 73–1–2, 73–1–4, 73–1–5, 73–1–6 and 52–1–1, which were more resistant than Stone at Riverside in the same year, showed no appreciable resistance under these conditions. On August 8, in the whole planting only 2 plants remained healthy; both of these were dwarfs. As in the previous year no variety was able to survive in the blight epidemic at Shafter. Thus under one set of conditions significant differences in the reaction of varieties were apparent, but under other and more severe conditions these differences were obliterated.

The degree of resistance has been estimated in cases where the evidence of resistance was considered to be well established. Table 5 was prepared from the data at Riverside in 1924 and 1925 using the method described above (p. 53). Thus in 1924 Stone was 50 per cent and Dwarf Champion 29 per cent blighted. The difference is 21 per cent and its standard error 6.1 per cent. The odds are 19 : 1 that the true difference equaled or exceeded 12 per cent, or in other words that Dwarf Champion was at least 24 per cent less susceptible than Stone. In 1924 selection 73–1 appeared to be 24 per cent less susceptible than its parent variety Santa Clara Canner. In 1925 the

* Dr. Weberbauer writes, "Die Pflanze wächst zwischen Lima und Ancon, am Meerestrand, auf steinigem und natürlich auch salzigem Boden. Bekanntlich giebt es in diesem Gebiet niemals wirklich Regen nur feine Nebel befeuchten das Land während die Monate Juni bis October."

† The writer is indebted to Dr. J. T. Rosa, University of California for these data.

most resistant of the five selections from it, namely 73–1–6, appeared to be 15 per cent less susceptible than Santa Clara Canner, indicating a loss rather than a gain in resistance by a second year of selection. A comparison of the reaction of Red Pear in 1924 and 1925 suggests that this variety may be characteristically more variable in its behavior than Dwarf Champion. No significant differences were found in the reaction of Dwarf Champion and Red Pear or of Dwarf Champion and the most resistant selections of 1924 and 1925. It is

TABLE 5

CALCULATED MEAN DIFFERENCES IN SUSCEPTIBILITY TO BLIGHT AT RIVERSIDE

	Year	Compared with	Observed difference per cent blighted.	Standard error of observed difference, per cent.	By 19 to 1 odds (one direction) mean difference exceeded this per cent.	On same basis of odds mean difference, stated as per cent less susceptible than check, exceeded this per cent.
Dwarf Champion	1924	Stone	21	6.1	12	24
Dwarf Champion	1925	Santa Clara Canner	20	5.6	11	27
Red Pear	1924	Stone	18	7.2	6	12
Red Pear	1925	Santa Clara Canner	25	5.6	16	39
Selection 73–1	1924	Santa Clara Canner	27	8.6	13	24
Selection 73–1–6	1925	Santa Clara Canner	17	6.5	6	15

evident that no variety or selection has shown a very high degree of resistance to blight. For the present it may be assumed that, in a moderate blight epidemic, for every four blighted plants in Stone or Santa Clara Canner not more than three would be found blighted in the resistant varieties.

TRIALS WITH PROGENIES OF HYBRIDS

At the beginning of the present study Dwarf Champion was the only variety which had been reported to be resistant. Since this is not a desirable commercial variety, because the fruit is rather small and too soft for commercial purposes, the study of blight resistance in hybrid progenies was clearly desirable. Accordingly F_1 and F_2 progenies between Dwarf Champion or Dwarf Aristocrat and three commercial varieties of standard habit, Norton, Santa Clara Canner and Globe, were planted at Riverside and Shafter in 1924 and

1925. The F_1 progenies were all standard in habit of growth, showing the well-known dominance of standard over dwarf. At Riverside for the whole season of 1924 the proportion blighted in two such families combined was about the same as in the check variety Stone.

TABLE 6

BLIGHT IN HYBRIDS BETWEEN STANDARD AND DWARF VARIETIES

	Year, place and period	Habit	Number recorded	Number blighted	Per cent blighted	Check variety	Compared with check variety	Probability that the difference from the check variety is *not* significant.
							χ^2	P
C46, C59b. Fl. Dwarf Aristocrat x Santa Clara Canner, and Dwarf Champion x Norton, combined.	1924, Riverside, whole season		35	21	60	Stone.....	1	.32*
C46, C59b. Fl. Dwarf Aristocrat x Santa Clara Canner, and Dwarf Champion x Norton, combined.	1924, Shafter, to June 3		62	39	63	Stone.....	0	1.00
C42-2b, C46-1b, C49-1a. F2. Norton x Dwarf Champion, Dwarf Aristocrat x Santa Clara Canner, Dwarf Aristocrat x Globe, combined.	1924, Riverside, whole season	Standard....	44	16	36	Standard....		
		Dwarf.....	24	2	8		5	.02
C58-1. F2. Dwarf Champion x Santa Clara Canner.	1925, Riverside, whole season	Standard....	79	32	40	Standard....		
		Dwarf.....	23	2	9		9	.003
C46-1b. F2. Dwarf Aristocrat x Santa Clara Canner.	1925, Riverside whole season	Standard....	37	11	30	Standard....		
		Dwarf	11	1	9		2	.16
C95-1. F2. Dwarf Aristocrat x Red Pear.	1925, Riverside, whole season	Standard....	75	13	17	Standard....		
		Dwarf.....	27	0	0		4	.04
C58-1, C46-1b, C95-1. F2. Progenies combined.	1925, Riverside, whole season	Standard ...	191	56	29	Standard....		
		Dwarf.....	61	3	5		15	.0001
C58-1. F2. Dwarf Champion x Santa Clara Canner.	1925, Shafter, to June 22	Standard....	150	149	99	Standard		Less than
		Dwarf.....	62	44	71		39	10^{-7}

* In the direction of greater susceptibility.

The same two families were planted at Shafter in the same year and up to June 3 also appeared to be about as susceptible as Stone (Table 6, C46, C59b). Apparently the blight resistance of the dwarfs is recessive. Three small F_2 families were planted at Riverside in 1924. Together they contained 44 standard and 24 dwarf plants* (Table 6). The plants of each F_2 population were set out indiscriminately so that standard and dwarf plants grew in the same row; this probably gives a more accurate comparison with reference to blight than if the two types had been planted in separate rows as with two different varieties. In their reaction to blight these families were not comparable with the check variety Stone, as they were planted a month later. Within the F_2 families the dwarf plants were less blighted than those of standard habit and it was decided to test this association with larger progenies in the following year.

At Riverside in 1925 three F_2 progenies were planted (Table 6). The same association between the dwarf character and resistance to blight was more or less pronounced in all of these, and in C58–1 there can be little doubt of its significance. Again, in F_2 progenies at Shafter in 1925 up to June 22, when blight was very prevalent, a similar difference was observed and the association therefore seemed to be well established. The indication of resistance shown by all three dwarf varieties tested (e.g., Table 2) is in harmony with this conclusion. The data are not sufficient to test the degree of association.

The dwarf and standard characters depend on a single pair of allelomorphic genes, *d* and *D* as Price and Drinkard[9] have shown, the dwarf character being a simple recessive to standard. If the association observed between the dwarf character and resistance is complete and if, as it appeared, resistance vanished together with the dwarf character in F_1 of dwarf $\times$ standard, it seems probable the resistance is in some way bound up with the dwarf character and that some quality peculiar to the dwarf plants better enables them to resist blight. If so, dwarfness and resistance have a common genetic basis in the dwarf gene *d*, or else the resistance of dwarfs depends on some gene or genes completely linked with the dwarf gene. Thus recent work by Lindstrom[7] indicates a complete linkage between the genes for smooth (i.e., not peach) skin and dwarf. But if resistance and dwarfness depend on distinct genes that are not completely linked, then some resistant standard and non-resistant dwarf plants should appear in F_2 as a result of crossing over, and the proportions of blighted plants among the F_2 standards and dwarfs

* The excess of dwarfs over the expected proportion of 25 per cent was due to trisomic inheritance in the family C49–1a.

should differ from those in respective standard and dwarf parental races. It is proposed to plant several F_3 progenies in the coming season. That resistance is not conferred exclusively by the dwarf gene is shown by the resistant character of Red Pear and of selections of standard habit. This fact appears to support the hypothesis of partial linkage. It may be, however, that resistance in these standard varieties depends on genes other than those which produce it in the dwarfs. If so, it may be possible by hydridization to breed types with increased resistance. This is being attempted.

Jones[5] has pointed out that incomplete linkage exists between the dwarf gene and the gene or genes which cause the constriction of fruit as in Red Pear. Further evidence of such linkage was provided by an F_2 population from Dwarf Aristocrat $\times$ Red Pear grown at Riverside in 1925 with the following result:

Standard		Dwarf	
Unconstricted	Constricted	Unconstricted	Constricted
46	21	27	0

These data also indicate that the constricted character behaves as a simple recessive. If dwarf and resistance to blight are partially linked and if resistance in Dwarf Aristocrat and Red Pear is of the same nature a similar linkage should exist between resistance to blight and constricted fruit. In this F_2 family there was not much blight and only 7 blighted plants fruited. Of these six were unconstricted and one constricted (pear) and the result was inconclusive.

Triploid tomato plants have recently been discovered (Lesley[6]) and among the progeny of these, a simple trisomic type has been obtained in which the extra chromosome appears to be that associated with the *d D* pair of genes. If the *d* gene confers resistance, a simple trisomic plant of the constitution *Ddd*, although it is predominantly standard in habit, might well be more resistant than a *dD* diploid, and a *ddd* trisomic plant more resistant than a *dd* (diploid) dwarf. It is possible that the reaction of these types might throw light on the genetic basis of blight resistance, especially if a reliable method of testing the reaction of single plants could be devised.

On the whole perhaps the evidence indicates that the resistance of Dwarf Champion and Dwarf Aristocrat in some way depends on their dwarf character, and therefore that their resistance to blight is a recessive character dependent on the dwarf gene.

SUMMARY

The reaction to western yellow blight of tomatoes, shown by various varieties and progenies of single plants selected for resistance and also by some hybrid populations has been tested in two different localities during three seasons. The incidence of the disease varied widely according to the season and place of trial. The standard deviation of the proportion blighted per row varies in different fields and may considerably exceed the standard deviation of random sampling. This emphasized the desirability of a suitable arrangement of replicate rows containing an adequate number of plants and of a uniform field. Against an attack of moderate severity in which about half of the plants of the check varieties Stone and Santa Clara Canner blighted in the whole season, Dwarf Champion, Red Pear and Dwarf Aristocrat showed a fair degree of resistance; they were probably at least 25 per cent less susceptible than the checks. They also showed resistance to a more severe attack if the first part of the blight period only was taken into account. The Globe variety appears somewhat resistant, while Norton is about as susceptible to blight as Stone. The currant tomato, *L. pimpinellifolium,* seemed to be somewhat more susceptible to blight than Santa Clara Canner. None of the varieties had sufficient resistance to survive an attack of extreme severity in the early part of the season. Three years' selection for blight resistance in the commercial variety Santa Clara Canner resulted in strains with resistance about equal to that of the dwarfs and Red Pear.

The resistant character of the dwarfs behaves as a recessive and appears to depend on the gene for dwarf or possibly on a gene or genes more or less closely linked with it. The reaction of Red Pear and the standard selection showed that resistance may be obtained without the gene for dwarf. If resistance is genetically of more than one kind, it may be possible by crossing to breed a variety with increased resistance to blight.

LITERATURE CITED

[1] Fisher, R. A.
1925. Statistical methods for research workers. 1–239, 6 tables. Oliver and Boyd, Edinburgh.

[2] Henderson, L. F.
1905. Idaho Agr. Exp. Sta. Ann. Rept. (1904) 28–32.

[3] Humphrey, H. B.
1914. Studies on the relation of certain species of Fusarium to the tomato blight of the Pacific Northwest. Washington Agr. Exp. Sta. Bul. **115**: 1–22. pl. 1–5.

[4] Hungerford, C. W.
1923. Report of Director. Idaho Agr. Exp. Sta. Bul. **131**: 51.

[5] Jones, D. F.
1917. Linkage in Lycopersicum. Amer. Nat. **51**: 608–621.

[6] Lesley, J. W., and Margaret M.
1925. Triploidy in the tomato. Science n.s. **61**: 208.

[7] Lindstrom, E. W.
1925. Inheritance in tomatoes. Genetics **10**: 305–317.

[8] McKay, M. B.
1921. Third crop pest and horticultural report. Oregon Agr. Exp. Sta. 174–178.

[9] Price, H. L., and A. W. Drinkard
1908. Inheritance in tomato hybrids. Virginia Agr. Exp. Sta. Bul. **177**: 17–53.

[10] Rogers, S. S.
1913. The culture of tomatoes in California with special reference to their diseases. California Agr. Exp. Sta. Bul. **239**: 591–617.

[11] Shapovalov, M.
1925. Ecological aspects of a pathological problem (western yellow blight of tomatoes). Ecology **6**: 241–259.

[12] ———
1925. High evaporation: a precursor and a concomitant of western yellow blight. Phytopathology **15**: 270–278.

[13] Pearson, Karl, Editor
1914. Tables for statisticians and biometricians. Cambridge University Press. LXXXIII, 1–143.

[14] Yaw, F. L.
1924. Report on survey of the canning tomato industry with suggestions for improvements. California Agr. Exp. Sta. Circular **280**: 1–30. fig. 1–12.

[15] Yule, G. U.
1922. An introduction to the theory of statistics, 6th ed. xv, 1–415. C. Griffin and Company, London.

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THE ABSORPTION OF IONS BY CITRUS AND WALNUT SEEDLINGS*

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I. INTRODUCTION

This paper is a discussion of experiments on the exchange of ions between solutions and citrus and walnut seedlings, and is intended to extend our knowledge of certain problems in nutrition. The highly important rôle of the ions entering the cells of a plant has long been recognized, but the dynamics of the process are far from being understood.

Previous papers from this laboratory have been concerned chiefly with the study of the effects of salts on the growth and composition of the plant; this paper will deal with the ionic exchange as affected by growing plants. Our work has dealt chiefly with citrus and walnut seedlings which we have found to be very favorable for experimentation.

II. TECHNIC OF HANDLING THE CULTURES

A. Citrus Seedling Cultures

The seedlings were germinated in moist sphagnum moss and allowed to grow until the roots were 5 to 10 cm. long. Before placing the seedlings in the cultures the testa was taken from the cotyledons; the roots were dipped momentarily in distilled water and inserted into holes in tin lids placed on beakers containing the culture solution

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(fig. 1). Since three seedlings were inserted in each perforation they were firmly held without the use of cotton, which tends to promote the growth of injurious fungi. The cultures were grown in a glasshouse and the portion of the roof directly over the cultures was protected by suitable paper to prevent contamination of the cultures by falling substances, especially calcium compounds. The volume of the culture solutions was kept approximately constant by frequent additions of distilled water. Two liters of solution were always prepared, one liter being used in the beakers and the remainder saved for analysis.

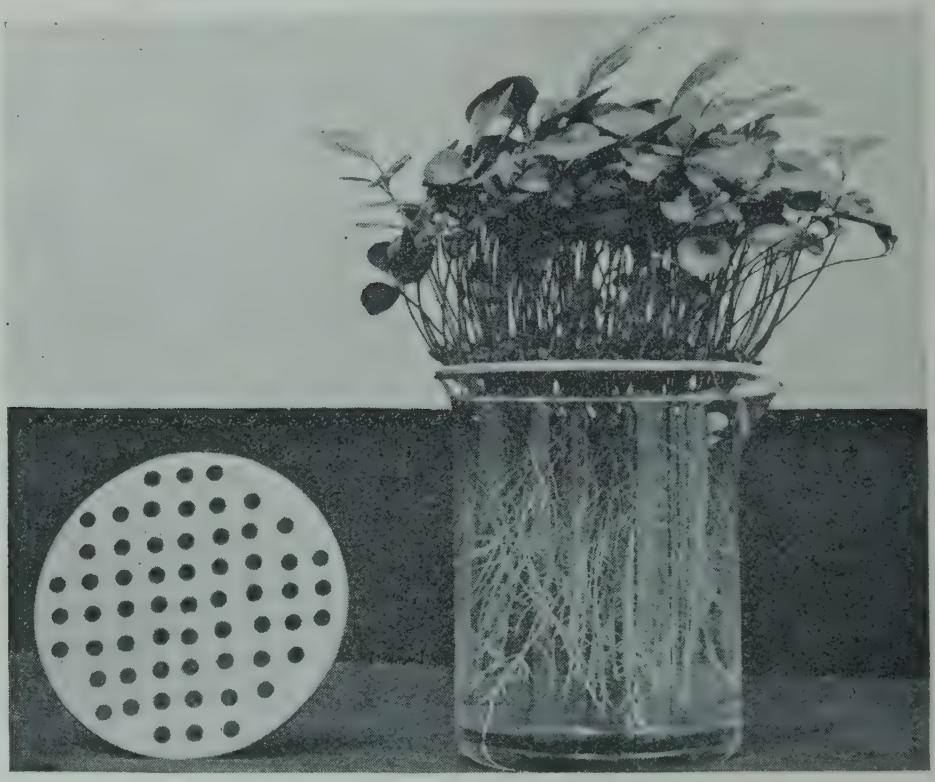

Fig. 1. One of the water cultures used for the study of absorption by citrus seedlings, with the perforated tin lid used shown at the left. The capacity of the beaker is one liter.

B. Walnut Seedling Cultures

Unbleached walnuts (*Juglans regia*) were placed in moist silica sand and kept in the glasshouse until the root just emerged from the shell (usually about four weeks), and then placed in moist sphagnum moss. When the tap root was 5 cm. or more in length, the seedlings were removed from the moss, the entire seedling (epicotyl not emerged as yet) was dipped momentarily in distilled water, and the excess water was blotted off by inverting the seedling and placing the shell against several sheets of filter paper. The roots are very sensitive to lack of moisture and if they become too dry the region just back

of the tip develops a light straw color, which disappears if the drying is not too prolonged. The seedlings grew well for several weeks in cultures of the type here described (fig. 2). Distilled water was added frequently to maintain the volume of the solution. In some cases it will be noted that the solutions were renewed, in others they were not changed.

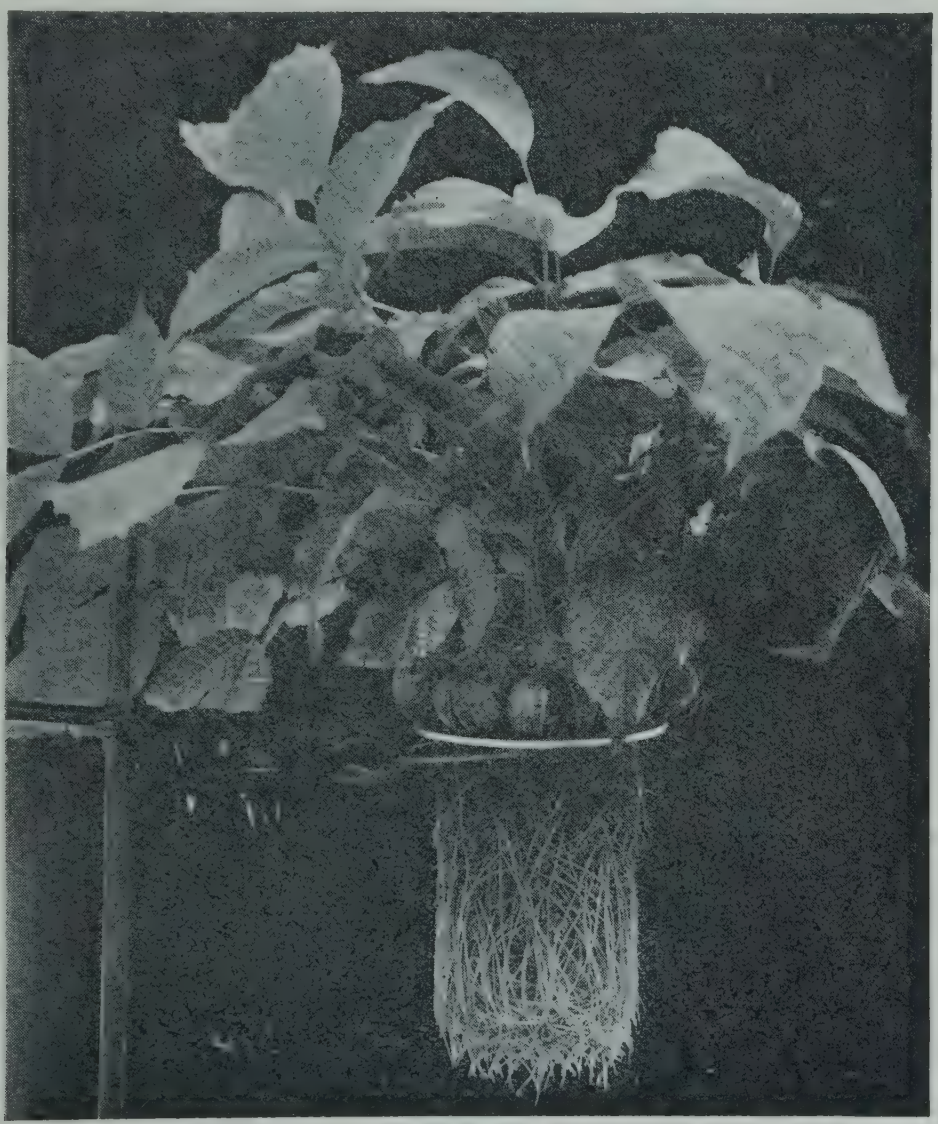

Fig. 2. A group of twelve walnut seedlings which had grown in a beaker like that shown in figure 1. Although the plants had been lifted from the solution, their matted roots held the form of the beaker in which they had grown.

III. EXPERIMENTAL DATA AND DISCUSSION

A. The Absorption of Ions by Citrus Seedlings

1. *The absorption of potassium ions.*

Five cultures of rough-lemon seedlings were grown in solutions which contained 7.7 milliequivalents K for 65 days and two cultures of grapefruit seedlings for 32 days (table 1). The cultures which contained KNO_3 and K_2SO_4 made the best growth, and those with

KH_2PO_4 were better than those with KCl. In the solution low in K, the plants made nearly as good growth as in the KCl set.

In two sets of cultures Cl or SO_4 was added only as KCl or K_2SO_4; thus the milliequivalents of K and anion are directly comparable since the anion was not furnished by any other salt.

TABLE 1

ABSORPTION OF IONS BY CITRUS SEEDLINGS FROM NUTRIENT SOLUTIONS CONTAINING EQUIVALENT AMOUNTS OF POTASSIUM

Salt added to nutrient	Reaction		Total milliequivalents of ions present							
	Initial pH	Final pH	Ca	Mg	Na	K	Cl	SO_4	NO_3	PO_4
A. Rough-lemon seedlings grown 65 days.										
KCl	5.5	4.7	8.323	4.786	1.337	7.613	8.483	5.244	7.734	3.369
K_2SO_4	5.5	5.2	8.114	4.754	1.215	7.726	0.525	13.243	7.499	3.709
KNO_3	5.2	5.7	8.083	4.901	1.484	7.828	0.606	5.312	14.659	2.950
KH_2PO_4	4.6	5.2	8.024	4.450	1.163	7.688	0.646	6.895	7.714	9.896
None	5.0	4.8	8.214	4.967	1.302	0.177	0.567	5.319	7.707	2.977
B. Grapefruit seedlings grown 32 days.										
KCl	7.0	4.8	7.934	4.934	1.866	7.770	8.139	4.905	7.253	1.435
K_2SO_4	7.0	4.8	8.263	4.721	1.662	7.816	4.740	7.885	6.973	1.542

Salt added to nutrient	Reaction		Milliequivalents of ions absorbed							
	Initial pH	Final pH	Ca	Mg	Na	K	Cl	SO_4	NO_3	PO_4
A. Rough-lemon seedlings grown 65 days.										
KCl	5.5	4.7	3.094	1.453	−.417	5.233	1.777	2.101	4.779	1.542
K_2SO_4	5.5	5.2	3.763	1.092	0.503	6.623	0.525	3.415	7.148	2.090
KNO_3	5.2	5.7	3.074	1.059	0.208	4.347	0.606	1.469	6.846	1.487
KH_2PO_4	4.6	5.2	3.114	0.649	−.551	4.887	−.164	1.373	6.054	lost
None	5.0	4.8	5.439	2.709	0.373	−0.046	−.322	1.808	6.438	2.142
B. Grapefruit seedlings grown 32 days.										
KCl	7.0	4.8	2.475	1.026	0.035	3.049	1.458	1.454	2.404	0.288
K_2SO_4	7.0	4.8	2.176	0.630	−.243	2.962	0.302	1.019	1.544	0.551

Table 1 shows that the reaction of the solutions was altered by the growth of the seedlings, but the shift was not always in the direction of neutrality. There was an increase in the acidity of the solutions which received KCl or K_2SO_4 and in that which received no K salt, while those which received KNO_3 or KH_2PO_4 became less acid.

Table 1 also shows the absorption of various ions, as determined by analyses at the end of the culture period. The amount of K

absorbed by the rough-lemon seedlings varied considerably but the quantities were not strictly correlated with the growth of the seedlings. A small amount of K was excreted by the roots where K was at a minimum. The amounts of Na absorbed were variable and there was no increased absorption of that ion where K was lacking. The plants absorbed more Ca and Mg when K ions were absent, but the total milliequivalents of kations absorbed was not greater than in the other solutions. The high rate of absorption of K by young seedlings is also shown in table 3. Unless a large amount of Ca was present, the seedling absorbed more K than Ca. These results are in agreement with those obtained by the ash analyses of orange trees grown in sand cultures (tables 12, 13, and 14). There was an increased absorption of the anion of the K salt where the larger amounts were present, although the plants in the K_2SO_4 set absorbed slightly more NO_3 than those in the KNO_3 set. These data as well as those of table 3, show that the citrus seedlings absorbed SO_4 and PO_4 ions in larger amounts than Cl ions, even when the initial concentration of the latter was considerably higher than that of either of the former. In every case except the KNO_3 series, the K absorbed exceeded that of the anion of the K salt. Where much K was supplied the absorption of Mg was greatest when KCl was added.

2. *The absorption of calcium ions.*

Several of our published papers have shown the importance of calcium and the large amounts found in citrus plants. We have made further studies on the absorption of ions from calcium solutions by seedlings of four species of citrus with reference to the reacting values of the ions absorbed. Table 2 shows that the solution became more acid as growth proceeded and that the plants absorbed more of the kations than of the anions that were furnished by the added calcium salt. The results agree with those of the preceding section in showing the greater absorption of the kation. These data show more or less significant differences in the amount of ions absorbed in different experiments. Cultures conducted during the months of March and April have usually produced the best plants and have shown the highest absorption of ions.

The addition of increasing amounts of Ca ions was reflected to a slight extent in the increased absorption, but the increases were by no means proportional to the amounts added. The data show that during certain periods the absorption of NO_3 by *C. limonia* was diminished by the addition of Cl ions. The original concentration of NO_3 in the four cultures was equivalent yet its absorption was lowered where $CaCl_2$ was added.

TABLE 2

THE ABSORPTION OF CALCIUM IONS BY CITRUS SEEDLINGS

Seedlings used	Duration of experiment	Reaction of culture solution		Calcium furnished (milliequivalents)	Milliequivalents of ions absorbed			
		Initial	Final		Ca	Cl	SO_4	NO_3
	Days	*pH*	*pH*					
Poncirus trifoliata Raf.	23	5.8	4.4	7.535	1.587	1.300		
Poncirus trifoliata Raf.	23	6.2	5.2	14.980	2.765	2.304		
Poncirus trifoliata Raf.	23	6.4	<5.0	22.360	2.919	2.622		
Citrus limonia Osbeck	65	6.0	4.0	7.779	4.845			3.864
Citrus limonia Osbeck	48	5.4	4.0	7.685	5.374			4.769
Citrus limonia Osbeck	48	5.7	4.0	15.369	6.013	1.523		3.434
Citrus limonia Osbeck	39	5.2	4.0	7.884	3.518			2.959
Citrus limonia Osbeck	39	5.4	4.0	15.429	3.533	0.818		2.038
C. maxima (Burm.) Merrill	36	5.0	4.0	7.794	2.745			2.346
C. aurantium Linn.	42	5.2	4.4	9.062	4.661			4.518
C. aurantium Linn.	46	6.0	3.8	26.766	10.080		8.776	
C. aurantium Linn.	46	5.2	4.3	7.884	4.491			4.358

3. *The absorption of chlorin ions.*

A series of experiments with rough-lemon seedlings was made to study the absorption of Cl from nutrient solutions to which different chlorids were added (table 3). Since these were added to nutrient

TABLE 3

ABSORPTION OF CHLORIDS BY ROUGH-LEMON SEEDLINGS

Chlorid added to nutrient solution	Duration of experiment	Reaction of culture solution		Milliequivalents of ions furnished							
		Initial	Final	Ca	Mg	Na	K	Cl	SO_4	NO_3	PO_4
	Days	*pH*	*pH*								
None	65	5.0	5.6	7.964	3.177	0.326	4.483	0.443	5.133		3.525
KCl	68	5.3	4.8	8.004	5.074	0.234	12.380	8.483	5.406	10.761	3.553
NaCl	68	5.3	5.3	8.184	5.148	6.662	5.445	8.282	5.206	10.980	3.553
$CaCl_2$	68	5.4	4.6	15.709	5.115	0.512	4.644	8.443	5.408	10.900	3.709
$MgCl_2$	68	5.2	4.7	8.383	12.077	0.456	4.700	8.564	5.418	10.760	3.501

Chlorid added to nutrient solution	Duration of experiment	Reaction of culture solution		Milliequivalents of ions absorbed							
		Initial	Final	Ca	Mg	Na	K	Cl	SO_4	NO_3	PO_4
	Days	*pH*	*pH*								
None	65	5.0	5.6	3.393	0.058	−.338	3.930	−.367	1.855		2.301
KCl	68	5.3	4.8	1.956	0.739	−.386	4.733	0.804	1.639	4.801	1.307
NaCl	68	5.3	5.3	2.335	1.305	0.868	3.154	0.606	1.217	5.319	1.307
$CaCl_2$	68	5.4	4.6	4.770	1.059	−.156	4.001	1.091	2.163	6 693	1.775
$MgCl_2$	68	5.2	4.7	2.136	0.993	−.226	2.496	0.567	1.206	3.718	1.096

solutions there was always a supply of the kation from other salts, except in the case where NaCl was the chlorid added. The amounts of Cl added were very nearly the same in the different cultures but after 68 days the analysis showed the greatest amount of Cl had been absorbed from the cultures which contained $CaCl_2$. This may be due to the fact that these plants made the best growth and hence were able to absorb more Cl without actually increasing its concentration in the tissues. The ratio of Ca to Cl in the solution at the outset was about 2:1, but the ratio of the milliequivalents absorbed was nearly 5:1.

Although the initial supplies of SO_4 and NO_3 were approximately the same in all cultures, the amounts absorbed were greatest in the solution to which $CaCl_2$ was added. This may be due to the favorable ratio between Ca and K in that solution. The absorption of PO_4 ions was greatest where chlorids were at a minimum.

The change in the reaction of the solutions showed the effect of differential absorption of anions and kations. In three cases the acidity developed was close to the limit of tolerance for citrus roots.

4. *The absorption of sodium and chlorin ions.*

With a greater concentration of NaCl than in the preceding experiment the results obtained with *Poncirus trifoliata* seedlings were somewhat different. The absorption of Na ions and that of Cl ions was practically equal (table 4) and the reaction of the solution changed from pH 4.8 to 5.8.

TABLE 4

THE ABSORPTION OF Na AND Cl BY 192 *Poncirus trifoliata* SEEDLINGS GROWN FOR 27 DAYS IN A LITER OF NUTRIENT SOLUTION PLUS APPROXIMATELY 1000 P.P.M. NaCl

	Initial concentration (milliequivalents)	Ions absorbed (milliequivalents)	pH
Na	16.544	2.513	
Cl	17.419	2.544	
Initial			4.8
Final			5.8

The relation of NaCl to the composition of young orange trees grown in sand cultures has been rather extensively discussed in previous papers. The leaves and shoots of such trees usually contained much more Cl than Na, especially if harmful amounts were employed, but in the trunks and roots Na was more abundant than Cl.

TABLE 5

REACTION CHANGES AND ION ABSORPTION INCIDENT TO THE GROWTH OF CITRUS SEEDLINGS IN CULTURE SOLUTIONS

	Chinese-lemon seedlings grown 35 days						Citron seedlings (72 seedlings to a liter) grown 27 days				Grapefruit seedlings (192 seedlings to a liter) grown 32 days			
	Hoagland's solution		Modified Hoagland's plus Na_2SO_4		Modified Hoagland's plus Na_2SO_4 and NaCl		Hoagland's plus 2×NaCl		Modified Hoagland's plus 2×Na_2SO_4		Modified Hoagland's plus KCl		Modified Hoagland's plus K_2SO_4	
	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed
	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.
Ca	7.804	2.186	8.114	1.906	8.134	1.826	8.104	1.038	8.034	1.088	7.934	2.475	8.263	2.176
Mg	5.008	1.026	4.860	0.624	4.860	0.624	4.844	0.714	4.860	0.534	4.934	1.026	4.721	0.632
Na	1.480	0.165	5.985	0.243	9.487	−.152	8.962	0.517	10.720	0.898	1.866	0.035	1.662	−.243
K	4.429	2.319	9.283	2.908	9.697	3.126	5.545	1.756	9.789	1.697	7.770	3.049	7.816	2.962
Total	18.721	5.696	28.242	5.681	32.178	5.576	27.455	4.025	33.403	4.217	22.504	6.585	22.462	5.770
						−.152								−.243
						5.424								5.527
Cl	0.485	−.040	0.183	−.181	4.526	0.406	9.069	0.767	0.282	−.121	8.139	1.458	4.740	0.302
HCO_3*	0.185	−.187	0.223	0.038	0.243	−.166	0.223	0.112	0.243	0.169	0.581	0.220	0.661	0.381
SO_4	5.495	1.352	7.072	1.302	7.045	1.206	5.514	0.840	12.944	1.105	4.905	1.454	7.885	1.019
PO_4	3.446	1.331	3.185	0.731	2.938	0.432	3.185	0.575	3.054	0.548	1.435	0.288	1.542	0.551
NO_3	9.597	2.826	18.834	4.589	18.753	3.887	10.219	1.703	17.451	1.523	7.253	2.404	6.973	1.544
Total	19.208	5.509	29.497	6.660	33.505	5.931	28.210	3.997	33.974	3.345	22.313	5.824	21.801	3.797
		−.227		−.181		−.166				−.121				
		5.282		6.479		5.765				3.224				
Initial pH	5.0		5.0		5.0		5.1		5.1		7.0		7.0	
Final pH	4.8		5.6		5.4		4.8		4.8		4.8		4.8	

* Determined by titration with methyl orange.

5. *The relation between ion exchange and reaction change of culture solutions.*

In table 5 we have included a few of the representative results of numerous experiments made on the changes which take place in culture solutions as a result of the growth of citrus seedlings. The concentration of the solutions was not excessive and the growth period was long enough for measurable absorption to take place. The cortical tissues of the root of walnut seedlings tend to slough away in time, while with young citrus seedlings this does not take place. Consequently with citrus seedlings grown for short periods there is very little opportunity for acids to arise from the decay of organic matter. The low concentrations employed permit of fair analytical accuracy.

The table shows in milliequivalents the initial concentration of the solution and the amount of absorption. In the first five cultures the sum of the anions originally present exceeded the sum of the kations originally present and the solutions were acid. The last two solutions were apparently neutral to phenol red although the ionic balance would indicate slight alkalinity. In the second and third cultures the final pH showed a decrease in acidity as a result of the growth of the plants. In both these cases the total milliequivalents of anions absorbed was greater than that of the kations absorbed and consequently more kations than anions remained in the solution, which caused greater alkalinity or decreased acidity. In each of the other cultures the final pH of the solutions showed an increase in acidity. In these cultures the totals show that a greater amount of kations was absorbed than of anions, leaving as a result more anions than kations in the solution, with a consequent increase in the acidity. The prevailing conception regarding pH changes in culture solutions is that acid solutions [$(NH_4)_2SO_4$ excepted] tend to change in the direction of neutrality or less acidity and that alkaline solutions tend to change in the direction of neutrality or greater acidity. Our results agree with those of Hoagland[5] in showing that, where no organic secretions enter in as factors, that the changes in reaction of culture solutions may be attributed directly to differential absorption of ions together with negative absorption (excretion) of certain ions.

Table 5 also shows that in the third and seventh cultures there was a negative absorption of Na ions even though in the third considerable Na was present in the solution. In the fourth culture the absorption of Na was approximately the same as that of Cl, and in the fifth culture that of Na approximately the same as that of SO_4 where the initial concentrations were approximately similar. In the second and third cultures, the absorption of Na was considerably less than that

of Cl or SO_4 combined; but when KCl or K_2SO_4 were used (sixth and seventh cultures) the K absorbed always exceeded the anion supplied with it, and this factor alone contributed much to the increased acidity of the solution.

The remarkable changes in reaction produced even by young seedlings are shown by an additional experiment with Florida Sour orange seedlings in which the pH changed from 6.8 to 4.0 in 12 days. A parallel set of cultures, in which the initial pH of 6.8 was changed to 5.5 by the addition of citric or tartaric acids, also brought the reaction to pH 4.0 in 12 days. As time went on the roots became

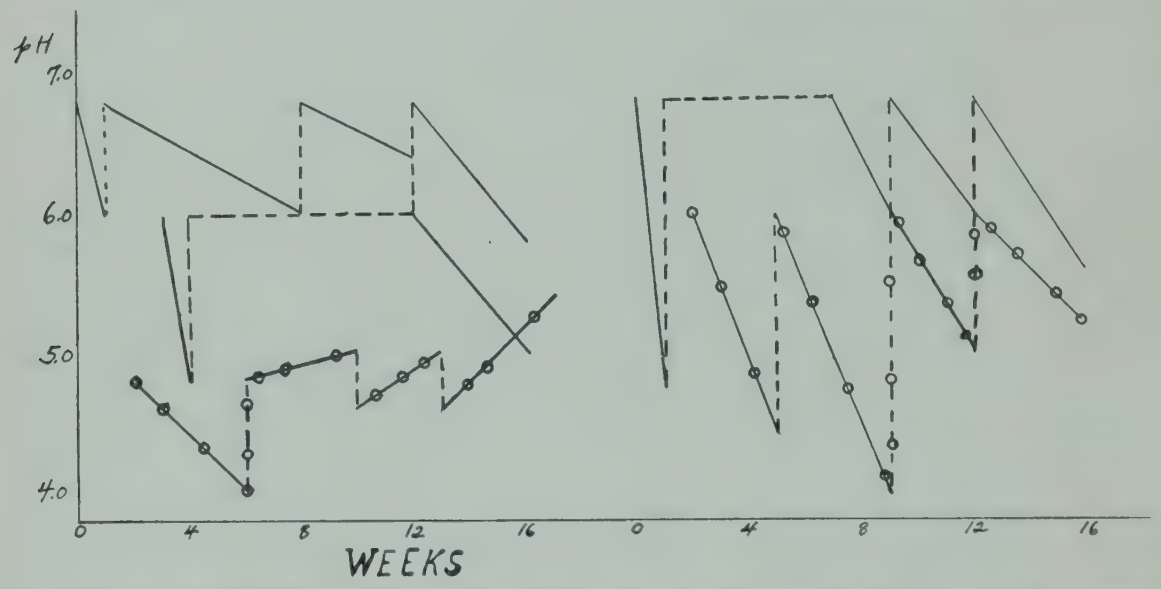

Fig. 3. The changes in reaction of culture solutions incident to the growth of citrus seedlings in them. ———— Sour orange; —o—o—o— Chinese lemon. Oblique lines represent changes brought about by the plants; vertical lines represent changes brought about by renewal of the solutions.

gelatinous but no growth of molds appeared. The results given in tables 4 and 5 show several cases in which the pH changes occurred in the opposite direction.

The changes in the reaction of culture solutions are shown by a series of graphs in figure 3. In one case the initial pH of the culture solution containing Sour orange seedlings was 6.8 and in the other 6.0. In both cases the subsequent changes were in the direction of greater acidity, although the solutions were renewed at frequent intervals. The desired initial reactions of the culture solutions were obtained by taking suitable proportions of KH_2PO_4 and K_2HPO_4.

It appeared that the rate of change of the pH was greatest in the initial stages of growth. The slope of the graphs showing the early changes is greater than that of those showing later stages. A similar effect has been observed in the case of walnut seedlings, especially so long as the cotyledons remained attached. The changes in reaction

of Chinese-lemon cultures started at pH 6.0 agreed with those of the Sour orange cultures. Although the culture started at pH 4.8 dropped to 4.0 in the early stage of growth, the subsequent changes in reaction were toward higher pH values.

Since the change of reaction of a nutrient solution depends upon the exchange of kations and anions, it seems impossible to make predictions of the direction in which the reaction will change, unless we know something about the absorption process during that period. In other words, we must recognize a state of equilibrium between the plant, the solution, and the gases of solution and atmosphere.

6. *The absorption of CO_3 by seedlings.*

Breazeale[2] has suggested that when nitrate is present as the anion in a single-salt solution, the plant may take up more NO_3 than of the kation, and that from the bicarbonate formed in the solution, the plant absorbs its supply of what becomes carbonate when the plant is ashed.

We have grown citrus seedlings in solutions of calcium nitrate and found that both the ash of the tops and that of the roots gave a strong effervescence with acid. Low heat was always used in ashing and the plants were not allowed to catch fire. The ash of tops of rough-lemon seedlings taken from $CaCl_2$ cultures as well as the ash of roots of grapefruit seedlings taken from $CaSO_4$ cultures in which the solutions had become either slightly acid or strongly so showed effervescence with acid. Analyses were made of control rough-lemon seedlings which had been grown in moss and whose outer seed coats had been removed prior to being ashed. The ash of these seedlings showed faint, if any, effervescence with acid. We have found that the ash of wheat seed gave no effervescence with acid. When wheat seed was treated with NaCl or $NaNO_3$ in dilute solutions, then dried and ashed, effervescence occurred with acid when ignition temperatures were high. When wheat was treated with dilute calcium nitrate solution, dried and ashed at various temperatures, effervescence always occurred with HCl. It appears therefore that the gentle ignition of any organic calcium compound will yield a carbonate. Hence the presence of CO_3 in plant ash is regulated largely by the relation between the absorption of kations and anions and volatility upon ignition.

7. *The behavior of citrus seedlings in sodium carbonate solutions.*

Breazeale[1] maintains that the toxicity of soil solutions containing small amounts of sodium carbonate is due largely to the action of the sodium carbonate upon the soluble organic matter. He found that

neither 400 p.p.m Na_2CO_3 nor a water extract of peat is toxic to citrus seedlings when taken singly but, when the two are mixed, the resulting solution is highly toxic.

In order to ascertain whether Na_2CO_3 up to 400 p.p.m. is stimulating, or at least non-toxic to citrus seedlings, we have grown grapefruit seedlings in cultures of carbon-treated distilled water containing 50, 100, 200, and 400 p.p.m. respectively of Na_2CO_3. The lowest concentration was between pH 8.5 and 10.0 while the others all had pH values above pH 10. After 2 to 3 days, the 400 p.p.m. culture was alkaline to phenolphthalein while those with lower strengths were not. The culture solutions were not renewed. Nine days later most of the seedlings had gelatinous roots.

The experiment was repeated, using Chinese-lemon seedlings and concentrations of Na_2CO_3 equal to 50, 150, and 400 p.p.m. In 6 days most of the roots had become gelatinous. In none of the cultures was there any evidence of elongation of the roots, and the high initial pH and the complete absence of Ca brought about gelatinization of the roots. The results indicate that the toxicity reported by Breazeale[1] may have been due to calcium starvation as well as to OH ions.

Soil containing black alkali, and soil which had been leached with NaCl until it was considered practically free from replaceable Ca, were leached with distilled water and the dark-colored leachate was collected. The solutions gave an alkaline reaction to phenolphthalein. They were then dialyzed against tap water until relatively free from Cl. The dialyzates were not alkaline to phenolphthalein. The dialyzed water-leachate of the soil containing normal carbonates contained 1826 p.p.m. total solids, with an ash content of 1132 p.p.m. The dialyzed water-leachate of the NaCl treated soil contained 2096 p.p.m. total solids and 1147 p.p.m. ash content. When citrus and walnut seedlings were placed in these dialyzed leachates, they grew very well, during the several weeks they were under observaton.

8. *The absorption by young orange trees.*

a. Water cultures. These experiments are largely of an exploratory nature, considerable difficulty having been experienced in the determination of suitable cultural methods. Sometimes trees taken from the field could be started in solution cultures but great difficulty was experienced in removing adhering soil. When all of the lateral roots were removed but not the tops, a fairly clean main root could be secured, but after several days the leaves wilted and the trees usually died. Where the leaves were removed, the buds frequently started but remained at a standstill for long periods.

TABLE 6A

Absorption During One Year by Young Valencia Orange Trees in Water Cultures

	Milliequivalents of ions in modified Hoagland's solution						Milliequivalents of ions in modified Hoagland's solution containing sodium chlorid			
	Tree 1		Tree 2		Tree 3		Tree 4		Tree 5	
	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed
Ca	255.039	59.077	286.078	129.760	285.104	102.734	281.147	78.388	281.212	78.453
Mg	108.413	11.133	104.062	22.060	94.456	4.442	105.293	9.130	102.141	5.230
K	150.129	31.585	140.053	44.060	147.121	25.224				
Cl							296.529	16.035	297.730	20.713
NO_3	323.346	113.310	343.461	182.654	348.045	226.381	355.325	140.502	359.505	120.670
SO_4	100.797	10.614	96.533	12.961	96.128	11.611	93.835	8.846	94.910	9.449
*PO_4	76.745	28.121	74.588	62.779	75.184	68.238				
Transpiration c.c.	31,525		46,115		37,125		41,770		33,000	
Initial pH	5.8		5.8		6.0		5 3		5.3	
Final pH	6.0		6.0		7.2		6.2		6.4	

* Cf. text, p. 81.

Budded Valencia orange trees secured from the nursery were placed in sand cultures after all branches, leaves, and rootlets had been removed. The top consisted of a piece of trunk about two feet in length and the root consisted of an undivided tap root about one foot in length. The root was scrubbed thoroughly with a brush to remove adhering soil. When such trees were placed directly into culture solutions a scum or film usually formed on the surface of the solutions. If the tree first regenerated some of its roots in sand cultures, such films seldom formed. After growing in sand kept moist with Hoagland's nutrient solution (from April until August 18), the trees were removed, the rootlets were freed from the sand and many of the rootlets were pruned away. The trees were then placed in 5-gallon wide-mouthed bottles containing a measured amount of nutrient solution, the partial composition of which is given in table 6A. Trees 1, 2, and 3 were placed in a complete nutrient solution which contained 318 p.p.m. Ca as calcium nitrate and 105 p.p.m. PO_4 derived from KH_2PO_4 and K_2HPO_4. Trees 4 and 5 received the same solution plus 297 m.e. Cl as NaCl. The culture bottles were kept in a box covered with boards and paper so as to exclude light, paper collars being placed about the trunks where they projected through the box cover. Distilled water containing iron was added from time to time but no additional nutrient was added from August 18, 1924, to September 28, 1925.

Fig. 4. Cultures used for the study of absorption by young orange trees after one years' growth made in bottles with a capacity of 20 liters.

The method of conducting the cultures and the type of the resulting growth are shown in figure 4. The trees produced about three cycles of growth on each of the larger shoots and the leaves were dark

green even though the cultures were grown in the glasshouse. There was some indication of mottle-leaf on the tree in culture 4 which received NaCl. The temperature of the glasshouse was continually maintained above 70° to 75° F and during certain periods of the year it was considerably higher. Some of the roots in the cultures receiving sodium salts died, and absorption by such root systems doubtless may be abnormal. The removal of PO_4 from the solution was due in a large measure to precipitation occasioned by the repeated additions of iron tartrate. The large volumes of water transpired by the trees are indicated in table 6A. The NO_3 absorption and the amount of transpiration furnish indication of the growth made by the various tree cultures. The absorption of NO_3 exceeded that of any other ion determined. Ca absorption, in some of the cultures that grew well (such as 2 and 3) was extremely large. It will be noted, however, that in proportion to the amount furnished, the absorption of K ranks close to that of Ca. The absorption of Mg was not very high, and the absorption of Cl and SO_4 ions was very low in comparison with that of the NO_3 ion. That larger amounts of anion than of kation were absorbed is indicated by the increased pH.

b. Sand cultures. Four sets of Florida Sour-orange seedlings were grown in sand cultures (10 plants to a 10-gallon crock) for about two years, by which time the plants had become approximately four feet high. The cultures received Hoagland's nutrient solution during the early period of development. The sand was then leached with distilled water until practically free from dissolved salts, and each culture was given four liters of complete nutrient solution, the composition of which is given in table 6B. After several weeks the cultures were again thoroughly leached and the per cent of the original ions absorbed was determined (see table 6B).

The nitrate absorption was greatest, practically all of the NO_3 being removed. The absorption of K and that of Ca were practically the same, while that of Mg was considerably less than either. The SO_4 absorption was less than that of PO_4 and NO_3.

9. *The relation of the H-ion to the growth of citrus seedlings.*

Previous experiments[9] on the absorption and growth of rough-lemon seedlings in solutions maintained at pH 6, 7, 8, and 9 showed optima at pH 8 and 9. We have made additional experiments with citrus seedlings and present herewith the results at pH 4, 5, 6, 7, 8, and 9.

TABLE 6B

ABSORPTION BY TWO-YEAR-OLD SEEDLINGS OF FLORIDA SOUR-ORANGE IN SAND CULTURES

	Absorption by two-year-old seedlings of Florida sour-orange in sand cultures		
	Milliequivalents furnished	Milliequivalents absorbed	Per cent absorbed
Ca	62.076	53.274	86
Mg	29.983	16.268	54
K	56.003	47.291	84
NO_3	71.999	70.678	98
SO_4	43.014	26.252	61
PO_4	29.009	20.309	70

Before suitable measurements of the effects of the H-ion concentration upon absorption by citrus seedlings can be adequately studied, we should first know the effects of H-ion concentration upon growth. The method of studying the effects upon growth has been usually that of starting with a certain nutrient solution and then regulating the pH of the culture solutions by the addition of suitable quantities of acid or alkali (usually H_2SO_4 and NaOH).

The present experiments make it evident that the initial pH of the nutrient solution adopted for such studies and the nature of the acid or base used in regulating the pH have considerable influence on the growth obtained at a given pH.

Grapefruit seedlings were grown in white-enamel pails containing 9 liters of Hoagland's solution of pH 5. One of the culture solutions was brought to pH 4 by the addition of HCl and the other solutions to pH 6, 7, 8, and 9 by the addition of NaOH. The pH of the solutions was readjusted two or three times daily and as frequently as changes in the pH of the solutions warranted. The solutions were renewed at intervals of from one to two weeks. Figure 5 shows the growth of plants from the different cultures after 43 days. The growth of the roots was least at pH 5.0. At pH 9 some of the leaves showed burning and some root tips were brown.

When St. Michael orange seedlings were grown in a similar manner for 56 days in Hoagland's solution whose reaction was brought to pH 4 by adding HCl and to 6, 7, 8, and 9 by adding NaOH, we found the poorest root growth again at pH 5 and the best growth at pH 7 with a definite decline at pH 9. In another experiment KH_2PO_4 was added to Hoagland's solution so as to give an initial reaction of pH 7. The pH was then adjusted by the addition of HCl and NaOH. Figure 6 shows the growth made in these solutions during

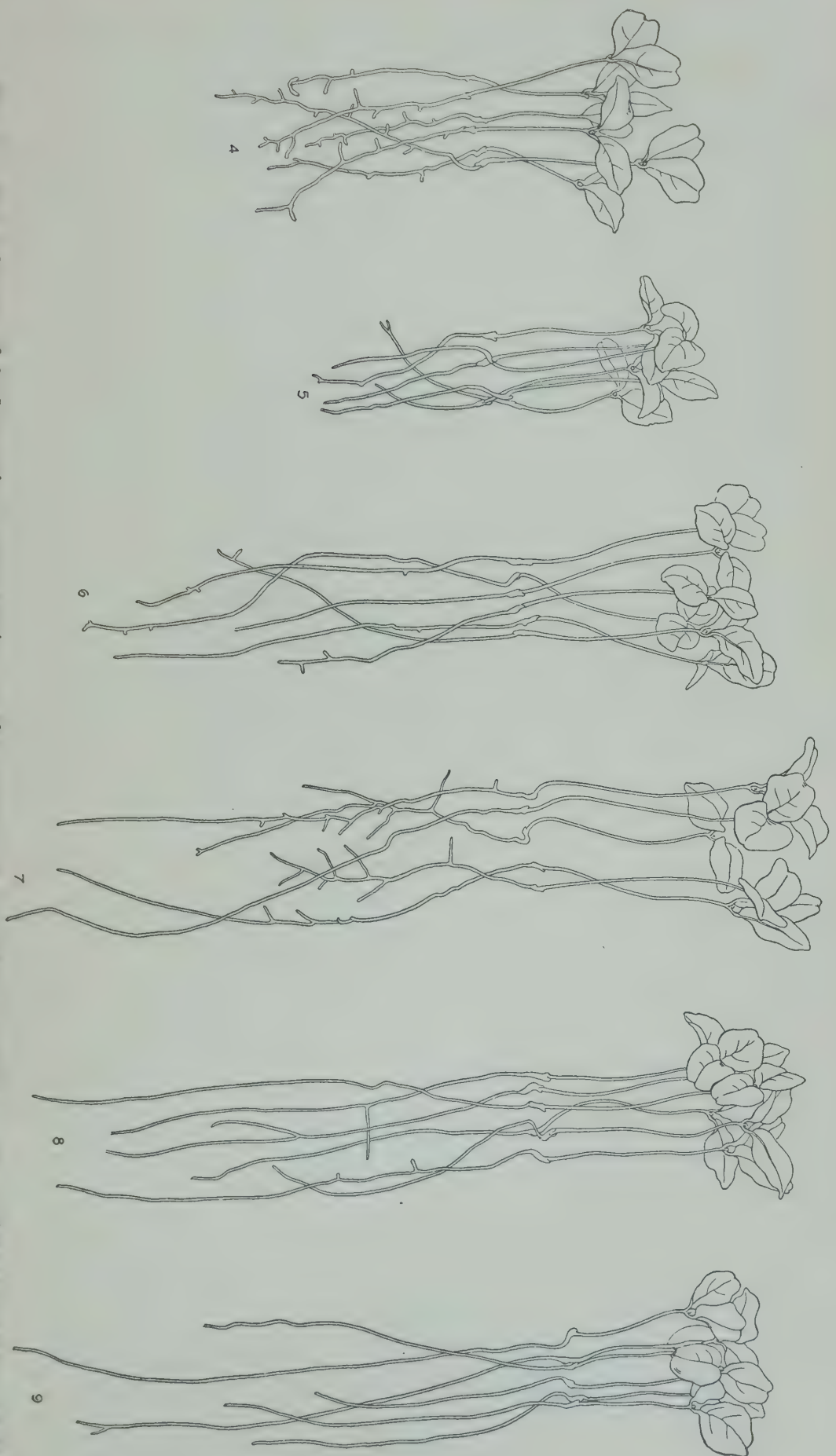

Fig. 5. The relation of hydrogen-ion concentration to the growth of grapefruit seedlings. The original reaction of the solution was pH 5.0 and was adjusted to reactions ranging from pH 4.0 to 9.0 by means of HCl or NaOH. The numerals indicate the pH values of the solutions.

Fig. 6. The relation of hydrogen-ion concentration to the growth of orange seedlings. The original reaction of the solution was pH 7.0 and was adjusted to reactions ranging from pH 4.0 to 9.0 by means of HCl or NaOH. The numerals indicate the pH values of the solutions.

a period of 45 days. The culture of pH 4 was no better than that at pH 5. The marked changes between the results at pH 6 and 7 and between those at pH 7 and 8 are evident.

Grapefruit seedlings were grown for 47 days in Hoagland's solution (reaction pH 5) and in Hoagland's solution regulated to pH 4 with $Ca(H_2PO_4)_2$ and to pH 6, 7, 8, and 9 with $Ca(OH)_2$. The poorest growth was made at pH 5, with greater growth at pH 4 and a marked increase at pH 6. If we use the solution which had an initial pH of 7, and change its reaction to pH 4, 5, and 6 with $Ca(H_2PO_4)_2$ and to pH 8 and 9, with $Ca(OH_2)$, we find a gradually decreasing length of the roots from pH 7 to pH 4 and excellent growth at pH 7, 8, and 9, with slightly the best at pH 8.

The results of these experiments emphasize some matters which have often been overlooked by previous investigators. They show the importance of (*a*) the hydrogen-ion concentration, (*b*) the original pH of the solution employed, and (*c*) the nutrient or toxic action of the reagents *per se* employed in maintaining the desired reaction. For example, the growth in a solution having an original reaction of pH 5.0 was improved for citrus growth either by decreasing or by increasing the hydrogen-ion concentration. On the other hand, growth in a solution having an original reaction of pH 7.0 was better than that in solutions having greater concentrations of hydrogen-ions. $Ca(OH)_2$ was much more favorable for growth than NaOH when used to increase the OH-ion concentration, as shown in a former paper.[9]

B. The Absorption of Ions by Walnut Seedlings

The problems of absorption were further studied by a series of experiments with walnut seedlings grown in culture solutions. On account of their extreme sensitiveness to certain kations walnut seedlings are well suited to the study of many problems akin to those already presented.

1. *The absorption from solutions containing a low concentration of potassium ions.*

The first series of cultures to be described was planned to study the absorption of nutrient ions when potassium was present in small amounts. The question was of no little importance for we have shown elsewhere[9] that walnut seedlings are relatively rich in potassium, and consequently may grow for some time without additional supplies of that element. The seedlings grew in the cultures for two periods of 35 days each. At the end of the first period of 35 days, the plants

were in a vigorous condition; consequently they were transferred to another similar solution and grown for a second period of 35 days, at the end of which time the seedlings were still healthy and vigorous. The roots were clear white, the leaves free from tip burn, and the solutions were free from the yellow tinge which often accompanies injury. The seedlings absorbed more of the anions and kations during the first period than during the second, with the exception of PO_4 which was completely absorbed during both periods (table 7). Although the ash of walnut kernels contains approximately 60 per cent of PO_4, the seedlings absorbed all of the PO_4 present in the solution. Although the culture solution contained a small amount of K at the beginning of the first period, no increase of K was evident at the close of that period. In the second period, however, when the initial concentration of K was lower, an excretion of K took place but no retardation of growth or unhealthy appearance of the plants due to lack of K was evident.

TABLE 7

ABSORPTION OF IONS BY WALNUT SEEDLINGS GROWN IN CULTURE SOLUTIONS LOW IN POTASSIUM

	First period (35 days)			Second period (35 days)		
	Original m.e.	Absorbed m.e.	Per cent absorbed	Original m.e.	Absorbed m.e.	Per cent absorbed
Ca	15.958	10.798	68	15.429	8.254	53
Mg	4.877	3.908	80	3.908	2.135	54
Na	3.355	1.484	44	1.167	−.100	.....
K	0.556	0.149	27	0.261	−.044	.....
Cl	0.606	0.606	100	0.364	−.059	.....
NO_3	14.381	12.519	88	14.381	9.412	65
SO_4	5.287	−.242	.....	5.273	0.478	9
PO_4	2.898	2.898	100	3.213	3.213	100

Additional results (table 8) were obtained from the two sets of cultures in which the absorption from complete and low K solutions was compared. The similarity in the per cent of ions exclusive of potassium absorbed in the two cases is remarkable.

The absorptions of ions by walnut seedlings differed from that of citrus seedlings when the concentration of K ions was low, in that no increased absorption of Ca or Mg took place (compare tables 1 and 8).

2. *The absorption of ions from solutions containing higher concentrations of potassium.*

The first two sets of cultures of walnut seedlings received Hoagland's solution, which contains 8 milliequivalents Ca (table 9). In

TABLE 8

ABSORPTIONS OF IONS BY WALNUT SEEDLINGS GROWN IN CULTURE SOLUTIONS FOR 39 DAYS

	Complete nutrient solution			Culture solution low in potassium		
	Original m.e.	Absorbed m.e.	Per cent absorbed	Original m.e.	Absorbed m.e.	Per cent absorbed
Ca	8.283	6.507	78	8.204	5.808	71
Mg	4.786	3.637	76	4.901	3.752	77
Na	1.016	0.109	10	1.098	−.373	.....
K	4.516	3.981	88	0.292	−.161	.....
Cl	0.558	0.319	57	0.558	0.358	64
SO_4	5.393	2.363	44	5.368	2.321	43
PO_4	3.473	3.473	100	3.054	3.054	100

cultures 3 and 4, and thereafter when high Ca is noted, we added 15 milliequivalents Ca as the nitrate in making up Hoagland's solution. Solutions 5 and 7 were the basis of a series in which the low K content was augmented by the addition of an appropriate K salt.

When the initial concentration of K was low, there frequently resulted an increase in the concentration of K in the solution. When

TABLE 9

ABSORPTION OF IONS BY WALNUT SEEDLINGS FROM SOLUTIONS CONTAINING HIGHER CONCENTRATIONS OF POTASSIUM

Culture	Modification of Hoagland's nutrient solution	Length of absorption period	Reaction of solution		Initial concentration of K	Ions absorbed	
			Initial	Final		K	Anion
		Days	*pH*	*pH*	m.e.	m.e.	m.e.
1	Nutrient solution (unmodified)	31	5.0	4.6	4.029	2.429	
2	Nutrient solution (unmodified)	39	5.4	4.6	4.516	3.981	
3	High Ca, first period	35	5.0	5.4	4.170	3.840	
4	High Ca, second period	34	5.3	6.0	4.288	3.320	
5	High Ca, low K, first period	35	4.8	7.0	0.556	0.149	
6	High Ca, low K, second period	34	4.9	6.4	0.261	−.044	
7	Low K	39	5.4	4.6	0.292	−.161	
8	High Ca, KCl, first period	35	5.0	5.5	7.839	6.743	2.442
9	High Ca, KCl, second period	34	5.3	6.0	7.839	3.817	1.334
10	KCl	30	5.3	4.6	7.808	7.483	2.183
11	KCl	39	5.2	4.6	47.265	22.339	14.083
12	High Ca, K_2SO_4, first period	35	4.9	5.5	7.790	6.687	2.949
13	High Ca, K_2SO_4, second period	34	5.0	6.0	7.662	4.787	1.909
14	K_2SO_4	30	5.2	5.2	7.734	4.009	3.251
15	K_2SO_4	38	5.0	4.4	39.683	14.121	13.347
16	High Ca, KNO_3, first period	35	5.0	5.6	7.880	5.717	15.083
17	High Ca, KNO_3, second period	34	4.9	6.3	7.759	4.063	14.342
18	KNO_3	30	5.0	6.0	7.767	4.429	8.301
19	KNO_3	39	5.4	6.2	86.502	26.225	
20	High Ca, KH_2PO_4, first period	35	4.6	5.2	7.823	5.985	3.967
21	High Ca, KH_2PO_4, second period	34	4.9	6.0	7.642	2.301	0.208

KCl was added to the culture solution, the absorption of Cl was always less than that of the K. The seedlings in cultures 8 and 9 removed less K from the solution containing 15 milliequivalents of Ca than from that containing 8 milliequivalents (culture 10). The most active absorption of K took place during the first period (culture 8). When the initial concentration of K was 47 milliequivalents a much smaller percentage absorption of K took place than when lower concentrations of K were employed, although the total amount absorbed was greater. The solutions used in cultures 1, 2, 7, 10, and 11 became more acid as a result of growth. Cultures 12 and 13 to which K_2SO_4 was added and which were high in calcium and nitrate were changed toward greater alkalinity, while those with lower Ca and NO_3 (solutions 14 and 15) tended toward greater acidity. There was less absorption of SO_4 than of K, even though the usual SO_4 of Hoagland's solution was present as well as that added with the potassium. Absorption was again greater in the first than in the second period. All the solutions used in these experiments contained calcium nitrate, consequently where KNO_3 was added the NO_3 absorption far exceeded that of the Cl, SO_4 or PO_4 anions. It is of interest therefore to find that, despite the large absorption of NO_3, the absorption of K in the first 35-day period was not greatly different from that of the cultures which received K_2SO_4. The reader may be reminded that a similar relation was found in the case of citrus seedlings (table 1).

The cultures which received KH_2PO_4 with high calcium nitrate showed a decrease in H-ion concentration and a greater absorption of K than of PO_4.

Table 10 gives the results of a determination of all ions (except NO_3) that were absorbed from solutions 11 and 15 of table 9. The milliequivalents of K absorbed in both cases were high, but were less than half the amounts initially present. From these data as well as from those in tables 9 and 13 it will be seen that less Cl and SO_4 were absorbed than K when a potassium salt was added in excess.

A series of cultures with walnut seedlings was conducted in which approximately 7.7 milliequivalents of K was added as a single salt to the nutrient solution. The solutions were analyzed and renewed at the end of 35 days. The plants were removed after growing 35 days in the second solution and the amounts of nutrient remaining in the solutions were again determined (table 11).

We found that, when large amounts of PO_4 were supplied, the total absorption for both periods was not greater than when the usual amount of PO_4 was supplied. It appears that where all the PO_4 was

TABLE 10

IONS ABSORBED BY WALNUT SEEDLINGS FROM TWO CULTURE SOLUTIONS CONTAINING AN EXCESS OF POTASSIUM SALT

	Initial concentration m.e.	Ions absorbed by plants m.e.	Per cent absorbed	Reaction	
				Initial pH	Final pH
Ca	8.134	3.423	42	5.2	4.6
Mg	4.754	1.995	42		
Na	0.074	−.903	.....		
K	47.265	22.339	47		
Cl	43.185	14.083	33		
SO_4	5.393	2.244	42		
PO_4	3.186	3.186	100		
Ca	8.144	2.894	36	5.0	4.4
Mg	5.222	1.314	25		
Na	1.332	0.933	70		
K	39.683	14.121	36		
Cl	0.502	0.169	34		
SO_4	48.458	13.347	28		
PO_4	3.342	3.342	100		

TABLE 11

ABSORPTION OF IONS BY WALNUT SEEDLINGS FROM SOLUTIONS CONTAINING EQUIVALENT AMOUNTS OF POTASSIUM

Modification of control culture solution	Period of 35 days	Initial milliequivalents of ions							
		Ca	Mg	Na	K	Cl	NO_3	SO_4	PO_4
Low K	First	15.958	4.877	3.355	0.556	0.606	14.381	5.287	2.898
Low K	Second	15.429	3.908	1.167	0.261	0.364	14.381	5.273	3.213
Plus KCl	First	15.689	4.770	1.697	7.839	8.483	14.628	5.231	2.769
Plus KCl	Second	15.549	4.951	1.424	7.839	8.364	14.628	5.183	2.977
Plus K_2SO_4	First	16.058	5.098	1.632	7.790	0.606	14.767	13.505	3.029
Plus K_2SO_4	Second	15.249	4.811	1.371	7.662	0.403	14.767	13.322	3.002
Plus KNO_3	First	16.048	4.770	1.580	7.880	0.809	22.237	5.115	2.977
Plus KNO_3	Second	15.469	4.918	3.333	7.759	0.567	22.237	5.260	2.662
Plus KH_2PO_4	First	15.788	4.967	1.463	7.823	0.606	14.683	6.725	18.045
Plus KH_2PO_4	Second	15.259	4.844	2.496	7.642	0.403	14.683	6.931	17.209

Modification of control culture solution	Period of 35 days	Milliequivalents of ions absorbed								Initial pH	Final pH
		Ca	Mg	Na	K	Cl	NO_3	SO_4	PO_4		
Low K	First	10.798	3.908	1.484	0.149	0.606	12.519	−.242	2.898	4.8	7.0
Low K	Second	8.254	2.135	−.100	−.044	−.059	9.412	0.478	3.213	4.9	6.4
Plus KCl	First	8.743	2.849	0.104	6.743	2.442	13.899	1.789	2.769	5.0	5.5
Plus KCl	Second	7.216	1.741	−.174	3.817	1.334	12.697	−.347	1.567	5.3	6.0
Plus K_2SO_4	First	8.822	3.021	−.517	6.687	0.606	14.339	2.949	3.029	4.9	5.5
Plus K_2SO_4	Second	6.587	1.905	−.195	4.787	0	8.005	1.909	3.002	5.0	6.0
Plus KNO_3	First	8.313	2.849	−.213	5.717	0.809	15.083	1.510	2.977	5.0	5.6
Plus KNO_3	Second	7.435	1.905	1.428	4.063	0.144	14.342	0.073	1.487	4.9	6.3
Plus KH_2PO_4	First	8.254	3.136	−.538	5.985	0.606	13.252	1.741	3.969	4.6	5.2
Plus KH_2PO_4	Second	8.513	1.486	0.937	2.301	−.203	11.759	−4.832	0.208	4.9	6.0

absorbed from the solution during the first period, the PO_4 constituted a limiting factor as indicated by the continued absorption during the second period. When the amount of PO_4 supplied was high, less K was absorbed during the second period. The total amount of Mg absorbed in each culture for both periods is quite constant. It is of interest to note that the absorption of SO_4 was negative in three cases and practically zero in a fourth. The nutrient solutions in every case were changed towards alkalinity. Except where KNO_3 was added, the K absorbed always exceeded that of the corresponding anion. The nutrient solution ordinarily used contained the primary potassium phosphate. Table 12 gives a comparison of this with solutions containing the secondary phosphate and a mixture of the two. The results are expressed as milliequivalents of ions absorbed by 12 walnut seedlings to a liter of solution during two periods of 33 and 49 days each.

TABLE 12

ABSORPTION OF K AND PO_4 IONS FROM NUTRIENT SOLUTIONS BY WALNUT SEEDLINGS

Potassium salt used in nutrient solution	Reaction			Initial concentration		Ions absorbed by plants		Per cent absorbed	
	Days	Initial	Final	K	PO_4	K	PO_4	K	PO_4
		pH	pH	m.e.	m.e.	m.e.	m.e.		
KH_2PO_4	33	5.0	4.6	4.355	3.081	3.937	3.081	90	100
KH_2PO_4	49	5.2	4.8	9.925	6.867	9.160	6.503	92	95
K_2HPO_4	33	7.0	4.6	5.123	2.690	4.575	2.690	89	100
K_2HPO_4	49	>7.0	4.9	10.985	6.111	10.084	5.588	92	92
K_2HPO_4 + KH_2PO_4	33	6.2	4.4	4.897	3.186	4.506	3.186	92	100
K_2HPO_4 + KH_2PO_4	49	5.6	4.7	10.143	5.248	8.940	4.544	88	87

It will be seen that the solution containing the primary phosphate had an initially lower pH value than the other two but that the final pH value in each case was very nearly the same. The amounts of K and of PO_4 absorbed were practically identical when expressed as percentages of the amount supplied. The large absorption capacity of walnut plants for PO_4, together with the great need of such plants for Ca, makes it desirable to test tricalcium phosphate as a source of Ca and PO_4. Such experiments are now under way but have not been completed yet.

3. *Absorption from solutions of calcium and potassium salts.*

Since it has not been possible to grow walnut seedlings in a solution lacking calcium, we made another experiment in which walnut seedlings were grown 47 days in solutions of KCl and of K_2SO_4

(approximately 7.7 milliequivalents of K) to which a small amount of $CaCO_3$ was added. The object of this experiment was to observe the absorption of K and its accompanying anion from solutions containing few other ions.

TABLE 13

ABSORPTION OF IONS BY WALNUT SEEDLINGS GROWN IN KCl OR K_2SO_4 SOLUTIONS CONTAINING $CaCO_3$

Solution	Initial concentration in milliequivalents				Milliequivalent ions absorbed				Final reaction
	Ca	K	Cl	SO_4	Ca	K	Cl	SO_4	pH
$KCl+CaCO_3$	12.475	7.716	7.930		7.595	6.077	2.121		5.5
$K_2SO_4+CaCO_3$	12.475	7.619		7.850	7.505	6.482		1.583	4.8

Table 13 shows that the seedlings absorbed approximately the same amounts of Ca from both solutions, and that although slightly more K was absorbed from the K_2SO_4 than from the KCl solutions, less SO_4 was absorbed than Cl. The results add evidence to that already presented showing the relatively rapid absorption of K by seedling plants.

Further studies were made on the absorption by seedlings of walnut and of St. Michael orange from solutions containing about 7.7 milliequivalents of K ions and about 8 milliequivalents of Ca ions. We have thus an opportunity to observe the absorption of these kations in the absence of all others and to compare the results with those where $CaCO_3$ was furnished.

TABLE 14

RELATIVE ABSORPTION OF K AND Ca IONS BY WALNUT AND ORANGE SEEDLINGS

Salts	Absorption period	Reaction		Initial concentration		Ions absorbed		Per cent absorbed	
		Initial	Final						
	Days	pH	pH	K	Ca	K	Ca	K	Ca
				m.e.	m.e.	m.e.	m.e.		
Walnut:									
$KCl+Ca(NO_3)_2$	32	4.8	4.4	7.424	7.744	2.726	1.996	37	26
$K_2SO_4+Ca(NO_3)_2$	32	5.0	4.6	7.342	8.044	2.696	2.774	37	34
Orange:									
$KCl+Ca(NO_3)_2$	32	5.0	4.6	7.557	7.924	3.190	2.335	42	29
$K_2SO_4+Ca(NO_3)_2$	32	4.6	4.8	6.774	7.984	2.952	3.094	44	39

Table 14 shows that the amounts of K and Ca absorbed were practically identical when the sulfate and nitrate were used, but when the chlorid and nitrate were used somewhat greater amounts of K were absorbed than Ca. In experiments (table 5) where complete nutrient

solutions were used and the initial concentration of K and Ca was approximately the same, the milliequivalents of K absorbed were greater than those of Ca. In table 14, however, we were dealing with incomplete culture solutions.

4. *The absorption of calcium by walnut seedlings.*

In a former paper,[8] we have shown the sensitiveness of walnut seedlings to the presence of calcium. We shall now give some data on the absorption from solutions of single calcium salts.

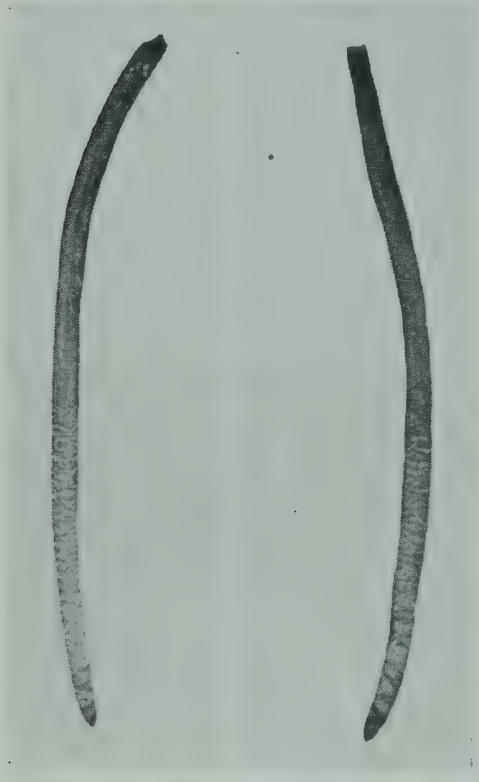

Fig. 7. Walnut-seedling roots showing the banded surface due to elongation after recovery from injury brought about by contact with a solution having unfavorable alkalinity.

The absorption of calcium from solutions of $CaCl_2$ was studied in two experiments (table 15), one of which ran for 20 and the other for 31 days. In each case the culture solution showed a slight increase in acidity due, as will be seen, to the greater removal of kations than anions, confirming the results obtained by Redfern.[6] In the 31-day period the plants absorbed about one-fourth of the calcium present and still less in the 20-day period.

The absorption of calcium from a solution of the primary phosphate (in spite of the higher acidity) was as great as from calcium chlorid solutions of equal strength. On the basis of the ions absorbed from the phosphate culture, we would expect the final reaction of the solution to become more acid instead of alkaline. We can account for the change in the opposite direction only by assuming that kations entered the solution from the plants. There was a certain amount of injury to the roots when first put into the calcium phosphate solution, but they later recovered only to show successive injury and recovery. This was particularly the case when 30 p.p.m. Ca as $Ca(H_2PO_4)_2$ was used. The root cortex of walnut seedlings usually darkens when the solution is markedly unfavorable. For example, if we place seedlings in $Ca(OH)_2$ solution of pH values in excess of pH 10, the root stops growing and the cortex becomes somewhat brown. If we then place the seedlings in a $Ca(OH)_2$ solution of more favorable pH, the root

begins to elongate, and as it does so the darkened cortex develops a banded appearance in the zone of elongation (fig. 7). Figure 8 shows the effect of $Ca(H_2PO_4)_2$ solutions upon the root tips of walnut seedlings grown in water cultures. When 30 p.p.m. Ca was used, the

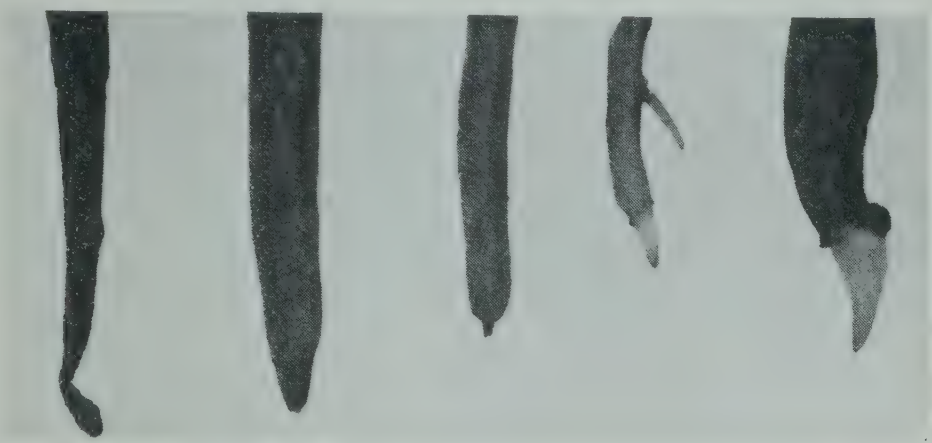

Fig. 8. Walnut-seedling roots showing injury and recovery. The three roots on the left were dead for a short distance back of the apex. The two roots at the right had regenerated new root tips and were beginning to recover.

changes in the effects brought about by this acid solution were readily followed. The roots died at the tip but not very far back. The cortical tissue then decayed away, leaving the central cylinder or stele still protruding. Gradually this disappeared and the root made another effort to grow from the apical end. Figure 9 shows successive injury and recovery and the broken-banded condition of the new roots similar to that shown in figure 7. The seedlings absorbed from a $CaSO_4$ solution about the same proportion of Ca as from the $CaCl_2$ solutions of equal concentration. The development of acidity in the $CaSO_4$ cultures is striking, especially in view of the relatively short duration of the absorption period in the first three cultures, where in some cases the epicotyls had scarcely emerged from the nuts at the end of the period.

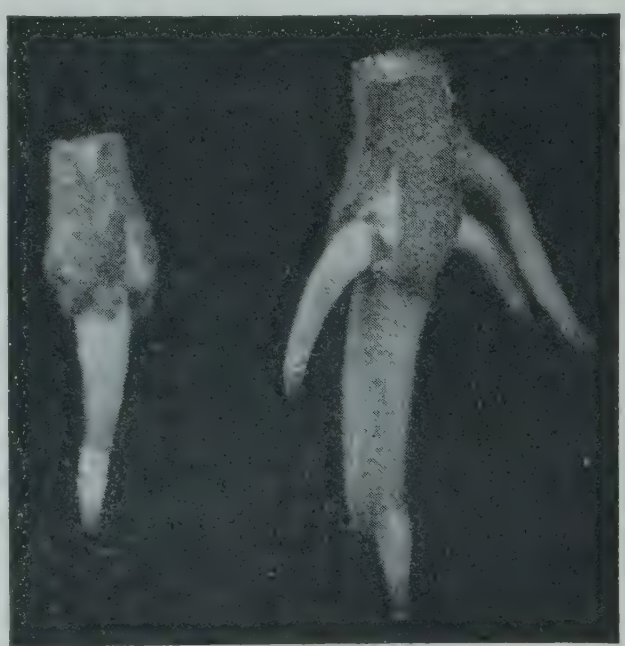

Fig. 9. Walnut-seedling roots showing successive injury and recovery incident to growth in a solution of $Ca(H_2PO_4)_2$. The way in which apical and lateral roots were formed from the stele indicates some difference in the vitality of the tissues of the roots.

The absorption of ions from solutions of calcium nitrate was considerably greater than from solutions of any other salts discussed in this connection. From the weakest solution the plants absorbed in a 20-day period 50 per cent and in a 47-day period 80 per cent of the calcium. The greater absorption of Ca in the case of the nitrate solutions is due in part to the more favorable final reactions of the culture solutions and in part to the greater absorption of NO_3.

Two other results should be noticed. The amount of the nitrate ion absorbed in these cultures was greater than that of the kation and the solutions decreased in acidity. In all the other cultures shown in this table the kation was absorbed in greater amounts than the anion, and as a result the solutions increased in acidity.

TABLE 15

ABSORPTION OF IONS BY WALNUT SEEDLINGS FROM SOLUTIONS OF VARIOUS CALCIUM SALTS

Twelve seedlings to a liter.

Salt	Initial concentration of Ca	Length of absorption period	Reaction		Ions absorbed by plant		Per cent of Ca absorbed
			Initial	Final	Ca	Anion	
	m.e.	days	pH	⅛ H	m.e.	m.e.	
$CaCl_2$	7.764	20	6.8	4.8	1.238	0.742	16
$CaCl_2$	15.100	20	7.8	4.8	2.096	1.689	14
$CaCl_2$	22.914	20	8.3	4.8	2.665	2.160	12
$CaCl_2$	7.685	31	5.4	<4.8	2.071	1.371	27
$CaCl_2$	15.838	31	5.8	<5.2	3.583	2.707	23
$CaCl_2$	23.463	31	5.6	<5.2	5.479	4.436	23
$Ca(H_2PO_4)_2$	7.575	23	3.8	4.2	1.637	0.652	22
$CaSO_4$	6.208	8	5.5	4.6	0.309	0.227	5
$CaSO_4$	12.250	8	5.8	4.8	0.494	0.198	4
$CaSO_4$	24.750	8	5.8	4.8	1.048	−.121	4
$CaSO_4$	20.928	20	5.4	4.0	3.124	1.874	15
$CaSO_4$	30.948	20	5.5	4.2	4.052	4.008	13
$Ca(NO_3)_2$	8.064	20	4.8	5.8	3.952	4.581	49
$Ca(NO_3)_2$	15.773	20	5.2	5.8	4.466	5.152	28
$Ca(NO_3)_2$	23.967	20	5.2	5.8	4.920	5.297	20
$Ca(NO_3)_2$	15.639	47	5.0	7.2	12.575	15.508	80

5. *The effect of other kations on the absorption of calcium by walnut seedlings.*

It has frequently been mentioned that the absorption of ions is modified by the presence of other ions. In their natural habitat the plant roots are in contact with many ions. It may, therefore, be interesting to note the effect of Na and K ions upon the absorption of Ca as shown in table 16.

The absorption of Ca from solutions of $CaCl_2$ was slightly diminished when NaCl or KCl salts were also present, although the absorp-

tion of Cl was increased. This diluting effect has been rather fully considered in other studies.[9] The addition of KCl or K_2SO_4 to $Ca(NO_3)_2$ solutions caused a final reaction more acid than the initial one, although when $Ca(NO_3)_2$ alone was used, there was a decrease in the acidity (table 15).

TABLE 16

THE EFFECT OF OTHER KATIONS ON THE ABSORPTION OF CALCIUM BY WALNUT SEEDLINGS

Culture solution	Initial concentration of Ca	Length of absorption period	Reaction		Ions absorbed		Per cent absorbed
			Initial	Final	Ca	Anion	
	m.e.	days	pH	pH	m.e.	m.e.	
$CaCl_2$	12.400	26	5.6	4.6	2.630	2.152	21
$CaCl_2$	24.880	26	6.0	4.6	3.453	2.829	14
$CaCl_2$+21.943 milliequivalents of Na as NaCl	12.744	26	5.4	4.6	2.126	3.706	17
$CaCl_2$+22.381 milliequivalents of Na as NaCl	25.050	26	6.0	4.7	3.024	4.746	12
$Ca(NO_3)_2$+7.424 milliequivalents of K as KCl	7.744	32	4.8	4.4	1.996	.....	26
$Ca(NO_3)_2$+7.342 milliequivalents of K as K_2SO_4	8.044	32	5.0	4.6	2.774	.....	34

6. *The passage of solutes from walnut seedlings into the solution.*

Walnut seedling roots soon die when maintained in pure distilled water or in calcium-free solutions.[8] On the contrary, they grow for some time if maintained in solutions of a calcium salt. If the seedlings with the cotyledons attached are kept for some time in moist moss or sand they make normal growth, limited only by the amount of material which the cotyledons can supply. The conditions responsible for the sudden injury to roots in distilled water, or in calcium-free solutions, form the pertinent starting point for an inquiry into the question of ionic exchange between the root and its environment.

Walnut seedlings are very sensitive to calcium salts, as was illustrated by an accidental introduction of a small amount of tap water into the distilled water which was being used. The roots made some growth whereas injury was soon apparent when the roots were kept in pure distilled water. It is remarkable that the harmful effects of distilled water upon walnut roots are so quickly evident. Within an hour the color of the sub-apical meristematic region of the root changes from healthy white to pale yellow. During the next few hours (depending somewhat upon the temperature) the sub-apical region changes from yellow to dark brown, probably due to the oxidation of a chromogen, which later appears to diffuse out of the roots, leaving them lighter colored.

While these color changes are occurring, the roots become gelatinous and eventually quite slimy for some distance back of the tip. The condition is altogether typical of that found in previous experiments[8] in calcium-free media. It has been shown that the gelatinous roots will produce laterals if they are transferred to suitable nutrient solutions before injury has involved the vascular system.

Some additional data are here given which bear upon the question of injury in pure distilled water. The experiment was a preliminary test to show whether the amount of ash constituents in a root is reduced when distilled water is used as a culture medium. One set was grown in nutrient solution, another in distilled water and a third in a saturated atmosphere. In distilled water there was opportunity for electrolytes to diffuse out of the roots. Only the seedlings in distilled water showed injury.

TABLE 17

Effect of Distilled Water on the Composition of Walnut Seedling Roots

	Control seedlings from damp moss	Seedlings after 3 days in distilled water
Fresh weight of 25 roots	25.00 grams	28.00 grams
Ash in the dry matter	7.12 per cent	6.40 per cent
Constituents of the ash:		
Na	4.65 per cent	3.20 per cent
K	26.77 per cent	17.39 per cent
Ca	.61 per cent	.36 per cent
Mg	1.07 per cent	.52 per cent
SO_4	3.65 per cent	1.72 per cent
PO_4	38.60 per cent	33.06 per cent

Twenty-five seedlings whose roots were 10 to 20 cm. long were placed in a culture jar containing 20 liters of carbon-treated distilled water. The roots from seedlings grown 3 days in distilled water and from seedlings taken directly from damp moss were cut just below the cotyledons, dried at 60° to 70° C and analyzed. Table 17 shows the composition of the roots of each set. The ash constituents of roots immersed for 3 days in distilled water were less than for those taken directly from the moss. The greatest relative difference was in the case of Ca, Mg, and SO_4. Our interest centers in the loss of Ca, because, as we have shown in another paper,[8] similar injury was noted when walnut seedlings were grown in nutrient solutions lacking Ca.

With our present knowledge of the salt requirements of plants it seems difficult to understand this Ca relation. In general there is no exact minimum requirement; on the contrary, as often shown, there is a more or less wide range in their salt requirements. One would be

slow to ascribe the sudden and profound effects observed to a change in Ca content from 0.6 per cent to 0.4 per cent. It is, of course, probable that there is a gradient for these ions in the roots, and that the diminution of Ca in the subapical region may have been much greater than that shown by the analysis of the whole root. The distribution of calcium in the walnut-seedling root was determined on a lot of roots which were about 12 cm. long when removed from the moss in which they had germinated. The roots were cut off immediately below the cotyledons and divided into basal, middle, and apical portions.

The calcium content of these roots is shown in table 18. The apical portion appears to contain more calcium than the others, although it is in this region that injury first appears when the roots are grown in calcium-free solutions.

TABLE 18

THE DISTRIBUTION OF CALCIUM IN THE ROOTS OF WALNUT SEEDLINGS DEPENDENT UPON THE COTYLEDONS

	Fresh weight	Dry weight	Per cent of Ca in	
			Fresh root	Dry matter
	(grams)	(grams)		
1. Basal part	43.68	4.89	.004	.038
2. Middle part	15.30	1.14	.008	.105
3. Apical part	7.87	.59	.009	.119

When walnut seedlings are grown several weeks in the usual nutrient solution the amount of Ca in the roots ranges from 3 to 5 per cent of the ash. Their total Ca content under favorable conditions for absorption is, therefore, not very large. Cranner[3] states that the presence of calcium is necessary to maintain the integrity of the bounding layer of the protoplast. In distilled water enough calcium may have diffused out of this layer to alter the permeability relations to a harmful degree without depleting to a corresponding extent the calcium content of the cell. Further study is necessary to determine whether the loss of this small amount of Ca from the walnut root is the primary cause of the injury noted.

7. *The absorption of sodium ions by walnut seedlings.*

A comparison of the ions absorbed from the nutrient solution and from the same with the addition of NaCl is given in table 19. In each case 12 walnut seedlings were grown for 39 days in 1 liter of solution. When walnut seedlings were grown in nutrient solutions

TABLE 19

ABSORPTION OF IONS FROM NUTRIENT SOLUTION WHEN SODIUM CHLORID WAS PRESENT

	Milliequivalents absorbed	
	Nutrient solution	Nutrient solution+87 m.e. of Na as NaCl
Ca	6.507	3.214
Mg	3.637	2.570
Na	0.109	12.538
K	3.981	3.295
Cl	0.319	12.574
SO_4	2.363	1.937
NO_3	.....	.....
PO_4	3.473	2.451
HCO_3*	.....	−2.56

* Determined by titration with methyl orange.

containing fairly large amounts of NaCl there was usually a characteristic suppression of growth in the epicotyls.

The results obtained from analyses of the residual solutions after the seedlings had grown for 39 days, show that the introduction of NaCl retarded the absorption of Ca more than that of the other kations. This effect has already been shown by the analysis of walnut roots grown in nutrient solutions containing sodium salts. We note also that Na and Cl were absorbed in molecular equivalents.

Additional experiments were made to determine the effect of NaCl in a more concentrated nutrient solution (table 20).

The concentration of the nutrient solutions was 1455 p.p.m. and 7275 p.p.m. respectively. Approximately 1275 p.p.m. of NaCl was added to one set and 2550 p.p.m to another. The four solutions in the order listed in table 20 had total concentrations, therefore, of 2730, 8550, 4005, and 9825 parts per million.

The roots of all seedlings in the four solutions showed no injury from the salts present, but the tops of the seedlings were affected. In solution No. 3 the margins of the leaves were killed in places. In solution No. 4 only two of the 12 seedlings put up epicotyls; the remainder were barely able to emerge from the shells. The plants which grew in the two weaker solutions (Nos. 1 and 2) were good in every respect.

The increased amounts of sodium apparently restricted the absorption of other kations. It seems evident from these data that the presence of an excess of NaCl operates to prevent the absorption of other ions, especially Ca. When the strength of the nutrient solution was increased fivefold there was no substantial difference in the amount of Na and Cl ions absorbed.

TABLE 20

ABSORPTION OF IONS BY WALNUT SEEDLINGS FROM NUTRIENT SOLUTIONS CONTAINING SODIUM SALTS

Twelve seedlings per liter were grown for 42 days.

Solution		Reaction		Initial concentration in milliequivalents						Ions absorbed by plants in milliequivalents						Ions gained by solution in milliequivalents
No.	Composition	Initial pH	Final pH	Na	K	Ca	Mg	Cl	SO_4	Na	K	Ca	Mg	Cl	SO_4	HCO_3*
1	Nutrient+1275 p.p.m. NaCl	5.4	4.8	19.717	6.633	8.234	7.192	22.219	.....	6.185	5.445	5.539	3.965	7.594	.....	0.148
2	(Nutrient×5)+1275 p.p.m. NaCl	5.0	6.5	23.314	24.389	39.022	25.476	23.189	.....	7.265	9.731	12.874	0.977	6.788	.....	3.049
3	Nutrient+2550 p.p.m. NaCl	5.4	6.5	39.598	10.268	7.754	7.430	42.820	.....	12.274	6.272	3.962	2.947	11.511	.....	1.207
4	(Nutrient×5)+2550 p.p.m. NaCl	5.0	6.5	43.196	24.781	39.571	25.024	45.185	.....	13.076	6.966	14.521	0.920	13.350	.....	2.453
5	Nutrient+640 p.p.m. Na_2SO_4	5.7	4.9	11.241	.....	15.569	8.612	.....	12.628	4.978	.....	10.130	5.451	.....	5.281	.....
6	Nutrient+1550 p.p.m. Na_2SO_4	4.9	5.3	22.178	.....	8.084	7.233	.....	26.121	5.981	.....	5.639	4.507	.....	5.042	.....
7	Nutrient+1550 p.p.m. Na_2SO_4 and 1275 p.p.m. NaCl	5.4	7.0	44.216	.....	8.184	7.299	21.029	25.599	6.380	.....	4.232	3.744	4.506	4.709	.....
8	Nutrient+3300 p.p.m. Na_2SO_4	4.9	5.0	41.451	.....	7.884	7.627	.....	49.758	11.701	.....	3.423	3.251	.....	9.108	.....

* Determined by titration with methyl orange.

When the amount of calcium was increased fivefold there was increased absorption, though not in proportion to the amount present. The case of magnesium appears somewhat unusual, because there was least absorption from solutions which contained most. It is possible that the absorption of calcium resulted in lowering the absorption of magnesium, which enters less readily into loose combination with the colloidal constituents of the cell. The diminished Mg absorption shown by solutions 5 to 8, accompanying increasing concentrations of Na ions, agrees with the reduced Ca absorption. The chlorin ion was absorbed in amounts substantially equivalent to those of its kation.

The effect of sodium sulphate was also studied (table 20). Some of the walnut leaves in solution 8 showed salt burn at the margins. The seedlings in solutions 5, 6, and 7 made excellent growth.

These results suggest that the roots have come into some sort of equilibrium with the ions in the solutions and that ions lost in the initial stage of the experiment may be absorbed later. The results reported by Hoagland[4] were obtained by placing a mass of roots of healthy plants into solutions for two days. In many cases he found a passage of ions from the plant to the solution where no doubt there would have been a net absorption in the course of time. However, our results agree in the main with those of Hoagland.

8. *The relation of the hydrogen-ion concentration to ionic interchange.*

In several of the foregoing experiments it was shown that the unequal rates of absorption of kations and anions resulted in changes in acidity. Our results agree with those of Hoagland[5] who concluded that the changes in reaction are due to unequal absorption and to the giving off of ions by the root. Many experiments have shown that the pH of the solution between pH 4.5 and 8.5 has practically no effect on the growth of walnut seedlings. This may be due to the large storage of reserve food within the seed. When the walnut plants grew in solutions well supplied with nitrate there was usually an accumulation of HCO_3 in the solution. When $Ca(NO_3)_2$ was furnished this effect was the more pronounced because of the rapid absorption of the NO_3 ions. The methyl orange titration of the nutrient solution calculated as HCO_3 was often as high as .7 milliequivalents at the end of a 33-day growth period and as high as 5.0 milliequivalents at the end of a second 33-day period.

The effect of ion interchanges is well shown by data in table 15. The net effect of absorption from $CaCl_2$ or $CaSO_4$ solutions was an increase in the H-ion concentration, while with absorption from $Ca(NO_3)_2$ solutions the OH-ion concentration increased. The pH of the $CaCl_2$ or $CaSO_4$ solutions usually reached 4.8 or 5.0 without evident

injury to the roots. When plants were put into solutions of $Ca(NO_3)_2$ growth continued for a longer time and pronounced increases in pH were observed. For example, the last set of cultures in table 15 grew for 47 days, during which time they absorbed most of the ions from the solution. The solution had a pH of 7.0–7.2 at the end of the period and upon heating to expel CO_2 it changed to pH 8.0. A small variation in pH, however, may be more significant in the more acidic solutions than is a large variation in the less acidic solutions.

C. General Discussion

When a living cell is in contact with a solution, absorption and excretion occur. The process never reaches an end-point because the ions absorbed by the plant promote its growth, thereby increasing its power or capacity further to absorb (or excrete) ions. It presents, therefore, a moving equilibrium. The exchange of ions is influenced by a great variety of conditions, such as the concentration of the solution, the nature and concentration of the ions present, the effect of one ion upon the absorption of another, temperature, light, and other factors. In fact, the absorption of ions is veiled by a host of factors, few of which are as yet understood.

Since energy is undoubtedly expended in the uptake of ions by plants, it is to be expected that the ratio and amount of this uptake should vary with the plant. The results herewith reported may therefore supplement those of Hoagland,[4] obtained from experiments with other plants. There is abundant evidence in this paper to support the idea that the processes of absorption are concerned with ions. The rate of absorption is not related to the velocities, nor to other physical-chemical properties of the ions so far as they are known. The amount of an ion absorbed is not strictly related to the amount already present in the plant. For example, phosphate was rapidly absorbed by walnut seedlings, although their cotyledons are very rich in phosphorus. We can only conclude therefore that their absorption is related to some chemical or physical property of the protoplasm.

Both citrus and walnut seedlings had a greater absorptive power for potassium, calcium, and nitrate ions than for magnesium, chlorin, or sulfate ions.

The effect of one ion upon the absorption of another was very pronounced, and occurred whether the ions had like or unlike electrical charges. Potassium ions retarded the absorption of calcium more than sodium, but sodium did not retard the absorption of

potassium. One of the striking features of these experiments is the rapidity with which potassium ions were absorbed. Whenever this ion was abundant it was absorbed, usually to the exclusion of other kations and sometimes anions.

In a soil where the rates of renewal and diffusion are slower than in a water culture there might not be such an accumulation of potassium in the plant. However, if the concentration of potassium were fairly high the absorption process might lead to exactly such a relation between potassium and calcium as we find in "mottle-leaf" and kindred troubles of citrus trees. There is evidence from data upon the absorption of ions by trees that the ratio of potassium to calcium absorbed normally becomes smaller with the growth of the tree, but the possibility of a later reversal and a return to this early condition exists.

Almost without exception the plants absorbed greater amounts of kation than anion, and the rate at which one was absorbed appeared to influence that of the other. The per cent of kations absorbed was always greatest when they were accompanied by a favorable anion like NO_3. The converse of this statement is generally true although the differences are not so striking.

The absorption of chlorin is of particular interest because of its occurrence in certain irrigated soils where it forms one of the constituents of "white alkali" salts. The detrimental effect of the absorption of any considerable amount of chlorin by plants has long been known. Our results indicate that the amounts of chlorin absorbed stand in the following order with respect to the accompanying kation: Ca>K>Na>Mg. It is probable that the larger amount absorbed is to a certain extent related to the growth promoting power of the individual kation. The larger plants thus produced have a large capacity to absorb ions. When barley plants of the same size were placed in the salt solutions the order of magnitude of chlorin absorbed was K>Na>Mg>Ca, according to Hoagland.[4] One of the effects of sodium chlorid upon the plant seems to be due to an interference with the absorption of calcium. As sodium chlorid accumulates in the leaves and meristematic regions it produces direct injuries, the nature of which is as yet unknown.

The nitrate ion was regularly absorbed in relatively large amounts even when the initial concentration was high. The presence of chlorin retarded the absorption of nitrate, but other ions seemed to have little effect upon its absorption. Where the initial concentration was sufficiently great the absorption of nitrate was sometimes greater than that of the corresponding kation. In the case of walnut seedlings,

the absorption of phosphate ions was also large, except where the concentration of the hydrogen ion was too great for satisfactory growth. It appears that the cells of the walnut root have some extraordinary powers of combining with phosphates, because when grown in distilled water there was no excretion of that ion, although the concentration of phosphorus compounds in the root was high.

In all cases studied there was an exchange of ions between the root and the solution in which it grew, resulting generally in a change in the acidity of the latter. The CO_2 excreted by roots tends to maintain a reaction favorable to growth. The most important effect noted was where the rapidly absorbed nitrate anion was furnished with a more slowly absorbed kation. In such a medium a fairly high degree of alkalinity may be developed by the formation of carbonates, but there was no evidence that any carbonate was absorbed by the roots.

The significance of the results of this study seems to lie in giving a clearer understanding of the processes of absorption from a relatively simple medium. From these results we have tried to obtain a clearer idea of the principles governing the process of absorption.

IV. SUMMARY

1. The data presented in this paper deal with the absorption of ions from solutions by citrus and walnut seedlings and by young trees. They deal with the relative amounts of various ions absorbed, the effects of one ion upon the absorption of another, and the changes in the reaction of the solution due to ion absorption and excretion.

2. Rough-lemon and grapefruit seedlings removed relatively more potassium than calcium from solutions containing approximately equivalent amounts of these ions. When the potassium in the culture solution was low in amount, citrus seedlings absorbed more calcium, magnesium, and phosphate from the solution than when potassium was abundant. There was an interchange of ions between the solution and roots resulting in an excretion of potassium into the solutions when the original concentration of potassium was low.

3. Citrus seedlings absorbed more kation than anion from solutions of single calcium salts, causing an increase in acidity of the solutions. Calcium ions were readily absorbed by citrus seedlings when sodium and potassium were absent or low in amount.

4. The presence of chlorin ions reduced the absorption of nitrate ions by citrus seedlings. When various chlorids were added to culture solutions the greatest amount of chlorin was absorbed from those containing calcium chlorid. These citrus seedlings made the best

growth and as a consequence were able to absorb more chlorin without injury than were the other cultures. From culture solutions which contained 1000 p.p.m. sodium chlorid, citrus seedlings absorbed approximately equivalent amounts of sodium and chlorin.

5. The changes in reaction of culture solutions in which citrus seedlings have grown, may be attributed directly to differential absorption of ions, together with an excretion of certain ions. Acid culture solutions do not always change in the direction of neutrality nor do neutral solutions always change in the direction of greater acidity. In complete nutrient solutions citrus seedlings may, in a comparatively short period, bring about so great a concentration of H ions as to be injurious to the roots.

Bicarbonate ions were found in culture solutions from which citrus roots had removed nitrate ions, but effervescence of the ash of citrus seedlings is not conclusive proof that the CO_3 had been absorbed from the solution.

6. No stimulation of the growth of citrus roots occurred in solutions of sodium carbonate. When calcium was completely absent from the solution, a high initial alkalinity due to sodium carbonate was very injurious to the roots of citrus seedlings; so also was the complete absence of calcium from a culture solution containing a favorable amount of the other essential ions.

7. Young Valencia orange trees removed more calcium than potassium from the culture solutions, but in comparison with the initial concentration the absorption of potassium ranked close to that of calcium. The absorption of magnesium was not very high in comparison with that of calcium and potassium. The absorption of the chlorin and the sulfate ions was very low in comparison with that of the nitrate ion, when the concentrations of chlorin and of sulfate were not excessive.

In sand cultures, Florida Sour-orange seedlings, two years of age, removed practically all of the nitrate present, and about equal percentages of the initial concentration of calcium and potassium. The percentage of the initial concentration of magnesium removed was much less than that of either calcium or potassium.

8. The nature of the growth obtained with citrus seedlings in water cultures at different pH values depends not only upon the maintained pH of the solution, but also upon the pH of the original solution as well as upon the nature of the acid or alkali used in maintaining the desired pH.

9. Although the ash of walnut kernels contains approximately 60 per cent of phosphate, the walnut seedlings rapidly absorbed all

of the phosphate from a complete nutrient solution. Walnut seedlings removed less chlorin or sulfate than potassium from culture solutions containing an excess of potassium salt. When walnut seedlings were grown in solutions having equivalent concentrations of potassium chlorid or potassium sulfate, they removed practically the same amounts of calcium from both solutions. Although slightly more potassium was absorbed from the potassium sulfate culture, the absorption of sulfate exceeded that of chlorin. Except in the case of potassium nitrate, the potassium absorbed by walnut seedlings from a nutrient solution to which different potassium salts were added, always exceeded that of the anion added with it.

10. The total amount of magnesium absorbed by walnut seedlings was quite constant and was not affected appreciably by additions of potassium salts to the solution.

11. Walnut seedlings removed more kation than anion from solutions of single calcium salts except in the case of the nitrate. The acidity of the residual solution was entirely dependent upon the absorption rate of the two ions employed. In a solution of calcium acid phosphate having an unfavorable degree of acidity, walnut seedling roots made renewed attempts to elongate, even after the root tips had been killed by the excessive acidity.

12. The addition of the chlorids or sulfates of sodium or potassium to calcium solutions brought about a reduction in the absorption of calcium. The addition of potassium chlorid or sulfate to calcium nitrate caused a final reaction more acid than the initial reaction, although when calcium nitrate alone was used, walnut seedlings brought about a decrease in acidity. The presence of an excess of sodium chlorid in a culture solution prevented walnut seedlings from absorbing large amounts of calcium. Increasing the concentration of the nutrient solution containing large amounts of sodium chlorid caused no substantial differences in the amounts of sodium and chlorin ions absorbed by walnut seedlings.

13. The apical portion of walnut roots grown in calcium-free solutions contained more calcium than the portions further removed from the apex, although the first evidences of injury were visible in this apical portion.

V. LITERATURE CITED

[1] Breazeale, J. F.
1919. Response of citrus seedlings in water cultures to salts and organic extracts. Jour. Agr. Res. **18**:267–274.

[2] Breazeale, J. F.
1923. The absorption of carbon by the roots of plants. Jour. Agr. Res. **26**:303–313.

[3] Cranner, Hansteen
1919. Beiträge zur Biochemie und physiologie der Zellwand und der plasmatischen grenzschichten. Ber. Deutsch. Bot. Ges. **37**:380–391.

[4] Hoagland, D. R.
1923. The absorption of ions by plants. Soil Sci. **16**:225–246.

[5] Hoagland, D. R.
1923. The effect of the plant on the reaction of the culture solution. Calif. Agr. Exp. Sta. Tech. Paper **12**:1–16.

[6] Redfern, Gladys M.
1922. On the absorption of ions by the roots of living plants. I. The absorption of the ions of calcium chloride by pea and maize. Ann. Bot. **36**:167–174.

[7] Reed, H. S. and Haas, A. R. C.
1923. Studies on the effects of sodium, potassium, and calcium on young orange trees. Calif. Agr. Exp. Sta. Tech. Paper **11**:1–32.

[8] Reed, H. S. and Haas, A. R. C.
1924. The effect of hydroxyl-ion concentration on the growth of walnut roots. Amer. Jour. Bot. **11**:78–84.

[9] Reed, H. S. and Haas, A. R. C.
1924. Nutrient and toxic effects of certain ions on citrus and walnut trees with especial reference to the concentration and pH of the medium. Calif. Agr. Exp. Sta. Tech. Paper **17**:1–75.

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THE INFLUENCE OF PRUNING ON THE GERMINABILITY OF POLLEN AND THE SET OF BERRIES IN VITIS VINIFERA

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In the progress of an investigation at the California Experiment Station of the effect of pruning on capacity, vigor, and bearing of Vinifera grapes,[1,2] it was observed that the type of pruning influenced the germinability of the pollen and the setting of the fruit.

So far as I have been able to find, no account of the influence of pruning on the germination of pollen has been published. This is also true with reference to the set of fruit, unless we except the many reports of larger yields resulting from the less severe or so-called "long" pruning in deciduous fruits. In this case, however, the larger yields which accrue from the development of a larger number of fruits may be the result of a larger bloom without any change in the quality of the flower parts, since the less severe pruning leaves a larger number of fruit buds on the tree.

Though there are no printed records of an increase in the set of fruits as a result of the long pruning of deciduous trees, the beneficial effect of blossom thinning on set has been indicated by Miss Bradbury.[1] She reports that during the one season of her tests, 1924, the set of fruits on sour cherry trees was increased from 24 per cent on the unthinned branches to 42 per cent on the branches on which the blossom buds of the spurs were thinned as early as practicable to one blossom to a bud.

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DEFINITION OF TYPES OF PRUNING USED

Normal Pruning.*—Pruning as nearly as possible in accord with the best accepted commercial practices of pruning each variety.[2] All bunches allowed to develop. (Fig. 1A.)

Severe Pruning.—Pruning similar to that of the normally pruned vines, but more severe, only the base buds being retained on the spurs. All bunches allowed to develop. (Fig. 1B.)

Half-long Pruning, part crop.—Pruning similar to that of the *normally pruned* vines, but less severe, six to ten buds being retained on the spurs. All bunches in excess of the number of bunches on the *normally pruned* vines, at the time of thinning, removed before blooming. (Fig. 1C.)

Cane Pruning, part crop.—Pruning similar to the commercial practice of *cane pruning* as used on Sultanina, except that more wood (four to nine canes, two to three feet long) is retained. All bunches in excess of the number on the *normally pruned* vines, at the time of thinning, removed before blooming. (Fig. 1D.)

No Pruning, part crop.—No pruning. All bunches in excess of the number on the *normally pruned* vines, at the time of thinning, removed before blooming. (Fig. 1E.)

No Pruning, all crop.—No pruning. All bunches allowed to develop. (Fig. 1E.)

THE GERMINATION OF POLLEN

Collection of Pollen.—Flower bunches under the different types of pruning were collected at as nearly the same time as possible on the same days and placed in bags. In the laboratory, the anther sacks were separated from the rest of the bunch and placed in vials. The pollen was tested for germination in no case later than 48 hours after its removal from the vine.

Germination.—In the germination tests, a small quantity of pollen was placed on a hanging drop of a sucrose medium in a Van Tieghem cell. In each test the number of germinated and ungerminated pollen grains in from ten to twenty areas over the surface of the drop was counted. Several hundred grains were counted for each variety under each type of pruning.

* Normal is used here in the sense of "usual."

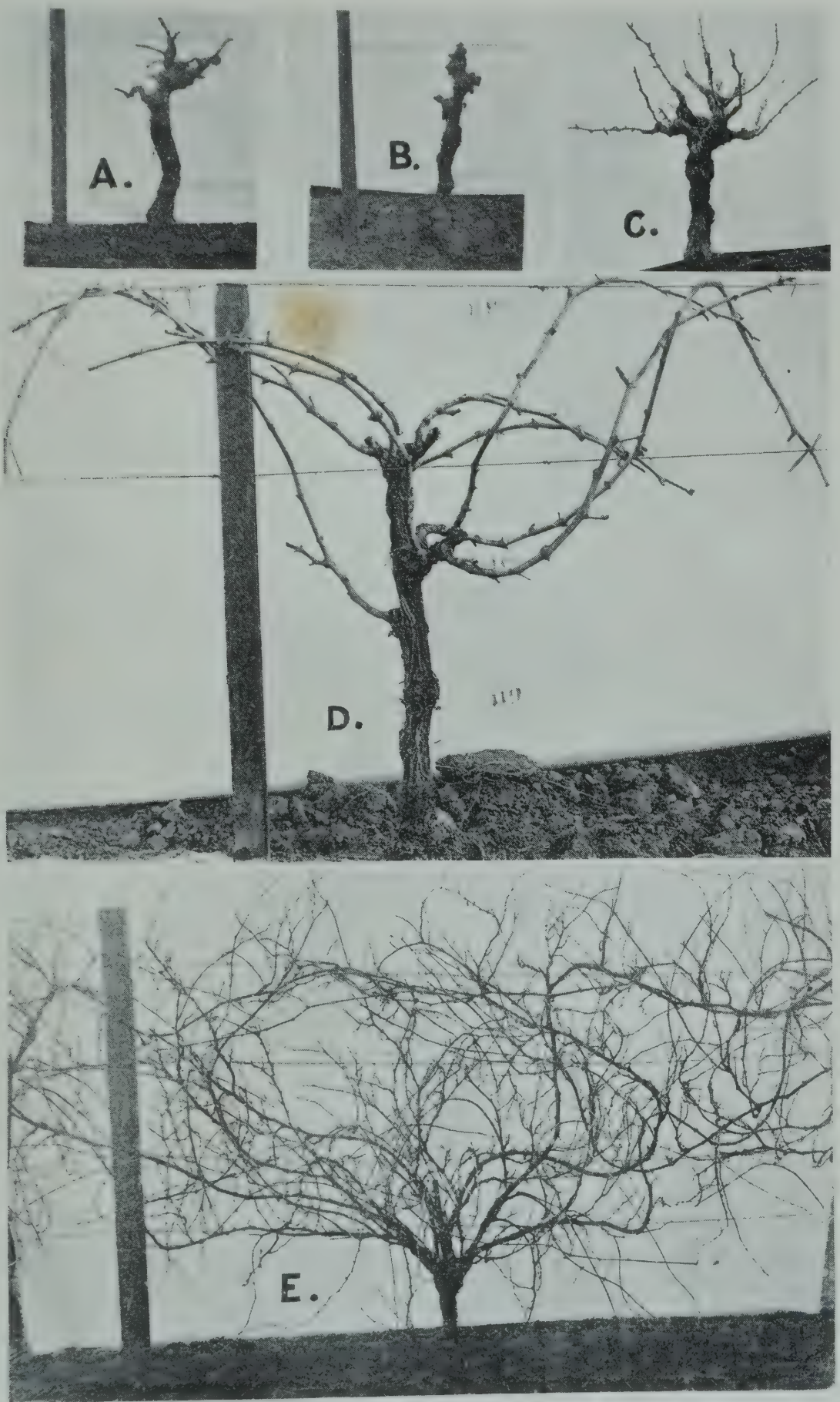

Fig. 1. Muscat of Alexandria vines showing the types of pruning used. A. *Normal (or usual) pruning.* B. *Severe pruning.* C. *Half-long pruning.* D. *Cane pruning.* E. *No pruning.*

The figures of table 1 indicate that grape pollen germinates well over a considerable range of temperature. Its sensitiveness to concentration of media, however, is more marked. Not a single grain germinated in water. In 15 and 20 per cent sucrose the germination was best, differing little in these two concentrations. Beyond these limits there was a rapid falling off in the germination.

In view of the data of table 1, all germination tests were made in both 15 and 20 per cent sucrose at 27°–30° C. In the table showing figures on germination, however, only the average per cent of germination for these two concentrations of media is given.

TABLE 1

THE INFLUENCE OF TEMPERATURE AND CONCENTRATION OF SUCROSE MEDIA ON THE GERMINATION OF GRAPE POLLEN

Temperature	Concentration of Sucrose Media—and per cent of germination						
	0 (Water)	5 per cent	10 per cent	15 per cent	20 per cent	25 per cent	30 per cent
18–21° C.	0	9	17	23	26	13	6
24–26° C.	0	9	17	27	33	17	9
30° C.	0	6	13	21	35	22	16
35° C.	0	4	7	20	27	24	17
Average	0	7	13.2	22.7	30.2	19	12

The samples of pollen of the Muscat of Alexandria, Black Monukka, and Alicante Bouschet were collected from the twelve vines under each type of pruning. For the other varieties the pollen was taken from five vines under each type of pruning. The pollen from all the vines of a variety under the same types of pruning was massed together, and after thorough mixing, was used as a single sample in the germination tests. The percentage of germination of the pollen of the several varieties under the different types of pruning is shown in table 2.

The data in table 2 indicate that the influence of pruning on the germinability of pollen is very marked. If we take the percentage of the germination of the pollen from the *normally pruned* vines as a standard, the germinability of the pollen of the *half-long pruned, part crop* vines was increased from 38 to 277 per cent; that of the *cane pruned, part crop* vines from 44 to 606 per cent; that of the *non-pruned, part crop* vines from 219 to 606 per cent; and that of the *non-pruned, all crop* vines from 117 to 576 per cent, when all of the varieties are treated as a whole.

The greatest increase was in Muscat of Alexandria and Alicante Bouschet, where the percentage of germination of the pollen from the *normally pruned* vines was very low. For the Muscat of Alexandria, for which the data for three years of the *non-pruned* vines are available, the average increase in germinability of pollen has been 277 per cent for the *half-long pruned, part crop;* 342 per cent for the *cane pruned, part crop;* 414 per cent for the *non-pruned, part crop;* and 296 per cent for the *non-pruned, all crop* vines.

TABLE 2

THE INFLUENCE OF PRUNING ON THE GERMINABILITY OF POLLEN

Variety	Year	Types of pruning—with per cent of germination					
		Severe	Normal	Half-long, part crop	Cane pruned, part crop	Non-pruned, part crop	Non-pruned, all crop
Muscat of Alexandria	1924	5.4	6.9	26	.....	26	15
Muscat of Alexandria	1925	7.8	8.0	.....	41	54	42
Muscat of Alexandria	1926	11.0	10.7	.....	41	52	45
Black Monukka	1925	17.3	17.6	46	.....	58	48
Black Monukka	1926	17.0	16.0	37	.....	51	40
Alicante Bouschet	1926	6.7	6.8	.....	48	48	56
Muscat gigas	1925	7.4	8.7	11	17	.....	.....
Muscat gigas	1926	5.0	12.0	17	19	.....	.....
Dizmar	1925	15.0	18.0	27	31	.....	.....
Dizmar	1926	15.0	15.0	.....	32	.....	.....
Molinera	1926	9	16	28	43	.....	.....
Henab	1925	33	37	58	57	.....	.....
Henab	1926	21	25	49	67	.....	.....
Malaga	1925	31	28	54	56	.....	.....
Malaga	1926	28	30	41	53	.....	.....
Emperor	1925	23	25	42	40	.....	.....
Emperor	1926	25	25	.....	36	.....	.....

In the case of such varieties as Malaga, Henab, and Emperor, where the pollen of the *normally pruned* vines gives a relatively high percentage of germination, the increase in germinability as a result of less severe pruning was not so great. It was, however, sufficient to be significant. The average increase in germinability of the Malaga pollen—which is typical of these varieties—was 65 per cent for the *half-long pruned, part crop* and 105 per cent for the *cane pruned, part crop* vines.

As might be expected, since the difference in the severity of pruning is relatively small, there has been no great difference between the germinability of the pollen of the *severely* and *normally pruned* vines.

The *severe pruning*, however, in twelve instances out of seventeen, decreased the germinability of pollen. This decrease varied from zero to 58 per cent. In two instances the percentage of germination was the same, and in three instances there was a very slight increase (2.5, 5.8 and 10.7 per cent).

THE SET OF BERRIES

It is the observation of growers wherever the Muscat of Alexandria and Hunisa are grown that these varieties are very subject to *Coulure* (shelling) and *millerandage*—the production of small seedless (shot) berries. The Muscat gigas is similarly defective with regard to *coulure*. These defects in the Muscat of Alexandria presented one of the first problems of the California grape growers to receive the attention of the Experiment Station. Efforts to overcome these varietal defects in California as well as in the other countries where these varieties are grown have been almost fruitless. In the other varieties listed in tables 3 and 4, these defects are rarely sufficiently serious to be of commercial importance. It is questionable, however, if there is a single variety of grape that under certain conditions of soil and climate is not subject to the setting of seedless berries or to shelling, and with many these defects are common.

In following up the influence of the different types of pruning on the set of berries, counts of the number of normal berries to a bunch were made. All of the berries on all of the bunches of six vines of Muscat of Alexandria, of four vines of Muscat gigas and of Molinera and of two vines each of Hunisa, Henab, Malaga, Emperor, and Ohanez under each type of pruning were counted. The numbers of normal berries to a bunch under the different types of pruning are given in table 3.

If the number of normal berries to a bunch on the *normally pruned* vines is taken as a standard, the data indicate an increase in Muscat of Alexandria of 221 and 312 per cent, respectively, for the years 1924 and 1925 in the number of normal berries to a bunch on the *non-pruned, part crop* vines. This increase in the case of the *non-pruned, all crop* vines was 57 and 68 per cent, respectively. In case of the *half-long pruned, part crop* and the *cane pruned, part crop* vines, the increase during 1925 was 114 and 238 per cent, respectively (fig. 2A and B).

In Hunisa, which of all the varieties of Vinifera grapes is one of the most prone to produce small seedless berries, the increase in the

number of normal berries was greatest. Here the increase was 407 per cent for the *half-long pruned, part crop* and 728 per cent for the *cane pruned, part crop* vines. In this variety the number of normal berries to a bunch was reduced almost to zero by *severe pruning.*

TABLE 3

THE INFLUENCE OF PRUNING ON THE NUMBER OF NORMAL BERRIES TO A BUNCH

Variety	Year	Types of pruning—and the number of normal berries to a bunch					
		Severe	Normal	Half-long, part crop	Cane pruned, part crop	Non-pruned, part crop	Non-pruned, all crop
Muscat of Alexandria	1924	32±1.6	37±1.1	77±1.9	.....	119±2.1	58±1.9
Muscat of Alexandria	1925	42±1.8	34±1.4	.....	115±3.6	140±2.4	57±2.9
Hunisa	1925	2±.31	14±1.8	71±5.0	102±9.1	.....	.....
Muscat gigas	1925	18±1.5	20±1.8	64±3.8	84±4.0	.....	.....
Molinera	1925	56±3.2	62±3.5	132±3.4	121±4.0	.....	.....
Henab	1925	80±4.2	80±5.0	109±7.1	123±6.1	.....	.....
Malaga	1925	103±13.2	132±14.0	183±7.0	166±11.0	.....	.....
Emperor	1925	100±7.3	138±6.8	164±8.7	165±8.9	.....	.....
Ohanez	1925	44±4.1	74±6.7	113±6.1	159±7.6	.....	.....

In Muscat gigas the increase in the number of normal berries to a bunch was 220 per cent for the *half-long pruned, part crop* and 320 per cent for the *cane pruned, part crop* vines. There was no significant difference in the number of normal berries to a bunch on *severely* and *normally pruned* vines.

In the other varieties—Molinera, Henab, Malaga, Emperor, and Ohanez—which are generally little affected by *coulure* and which usually set a very high percentage of normal berries, the number of normal berries to a bunch has not been so much influenced by the type of pruning. The increase as a result of the less severe pruning has, nevertheless, been considerable. The increase in the number of normal berries to a bunch has ranged from 19 to 112 per cent for the *half-long pruned, part crop* and from 20 to 115 per cent for the *cane pruned, part crop* vines. In these varieties the decrease in the number of normal berries to a bunch on the *severely pruned* vines also was small. The decrease ranged from zero in Henab to 40 per cent in Ohanez.

Of only slightly less importance than the number is the percentage of normal berries to a bunch. An increase in the number of normal berries to a bunch would not improve the quality of a grape or increase

its salability if it were accompanied by a corresponding increase of small seedless (shot) berries. That is, a large bunch of equally poor quality is little or no more desirable than a small bunch.

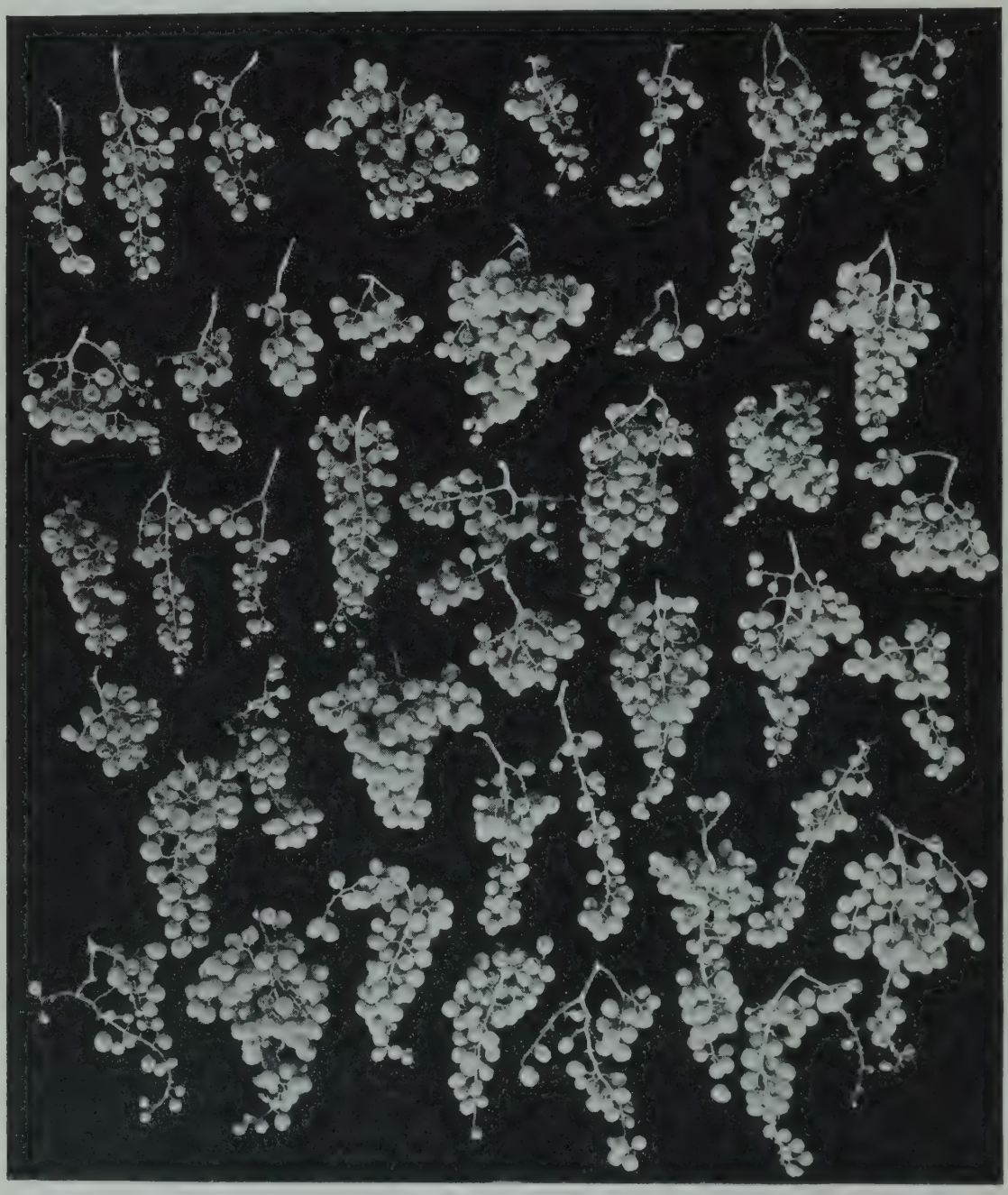

Fig. 2A. The influence of the type of pruning on the number of normal berries set to a bunch on Muscat of Alexandria. *Normal pruning* (34 normal berries to a bunch). (Entire crop from one average vine.)

At the time of the count of normal berries the total number of berries on each bunch was determined. The average percentage of normal berries to a bunch was then calculated from these counts. The percentage of normal berries to a bunch under the different types of pruning are given in table 4.

Again, if we take the percentage of normal berries to a bunch on the *normally pruned* vines as a standard, the average increase of normal berries to a bunch in Muscat of Alexandria has been 25 per cent for the *non-pruned, all crop;* 40 per cent for the *non-pruned,*

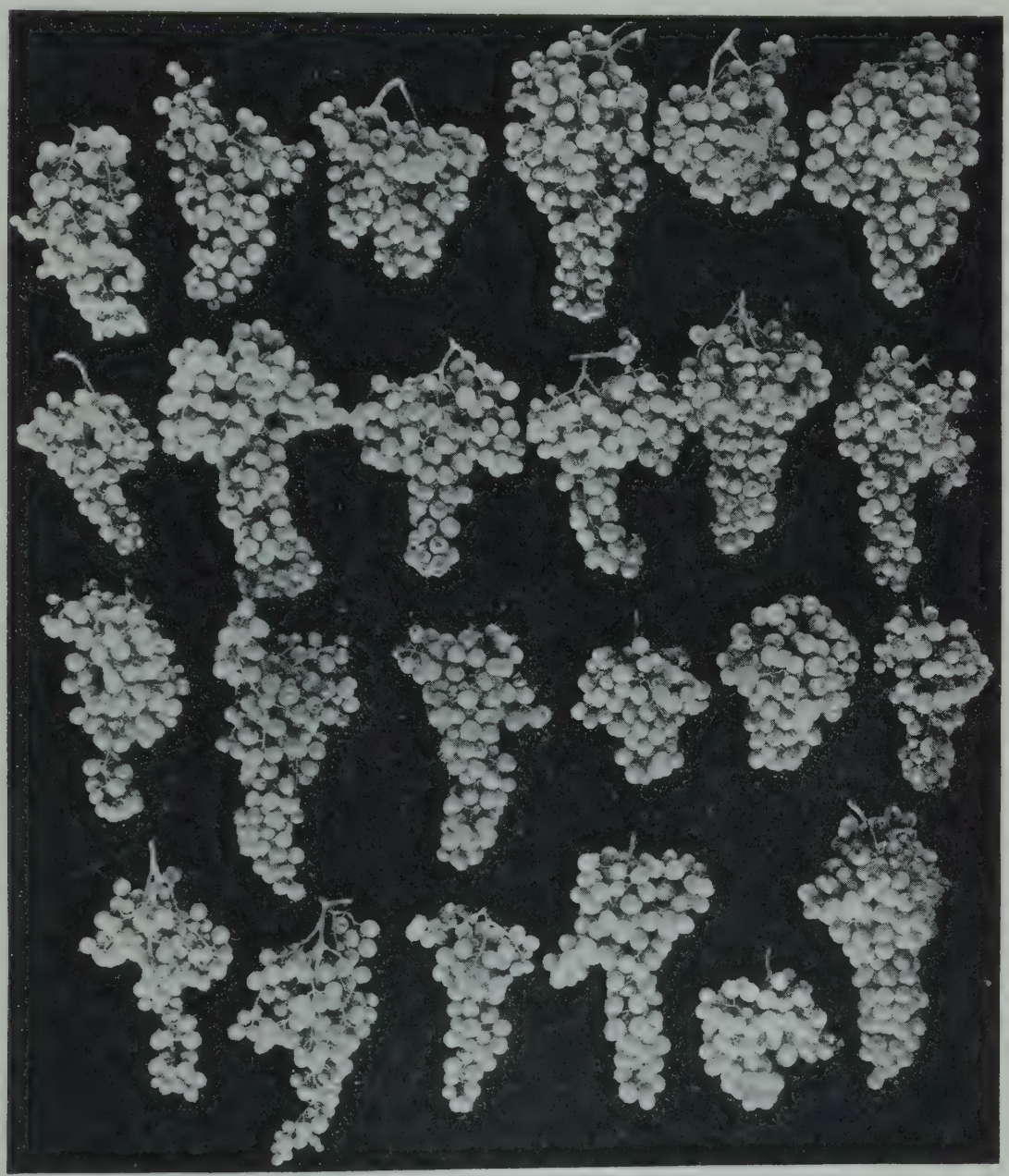

Fig. 2B. The influence of the type of pruning on the number of normal berries set to a bunch on Muscat of Alexandria. *Cane pruning, part crop* (115 normal berries to a bunch). (Entire crop from one average vine.)

part crop; and 36 per cent for the *half-long* and *cane pruned, part crop* vines. For Hunisa the increase has been very marked, being 230 per cent for *half-long pruned, part crop* and 590 per cent for the *cane pruned, part crop vines.* (Fig. 3A and B.)

The *severe pruning* in the case of these varieties also reduced the percentage of normal berries considerably. With the Muscat of Alexandria, the reduction was 18 per cent and with Hunisa, 75 per cent.

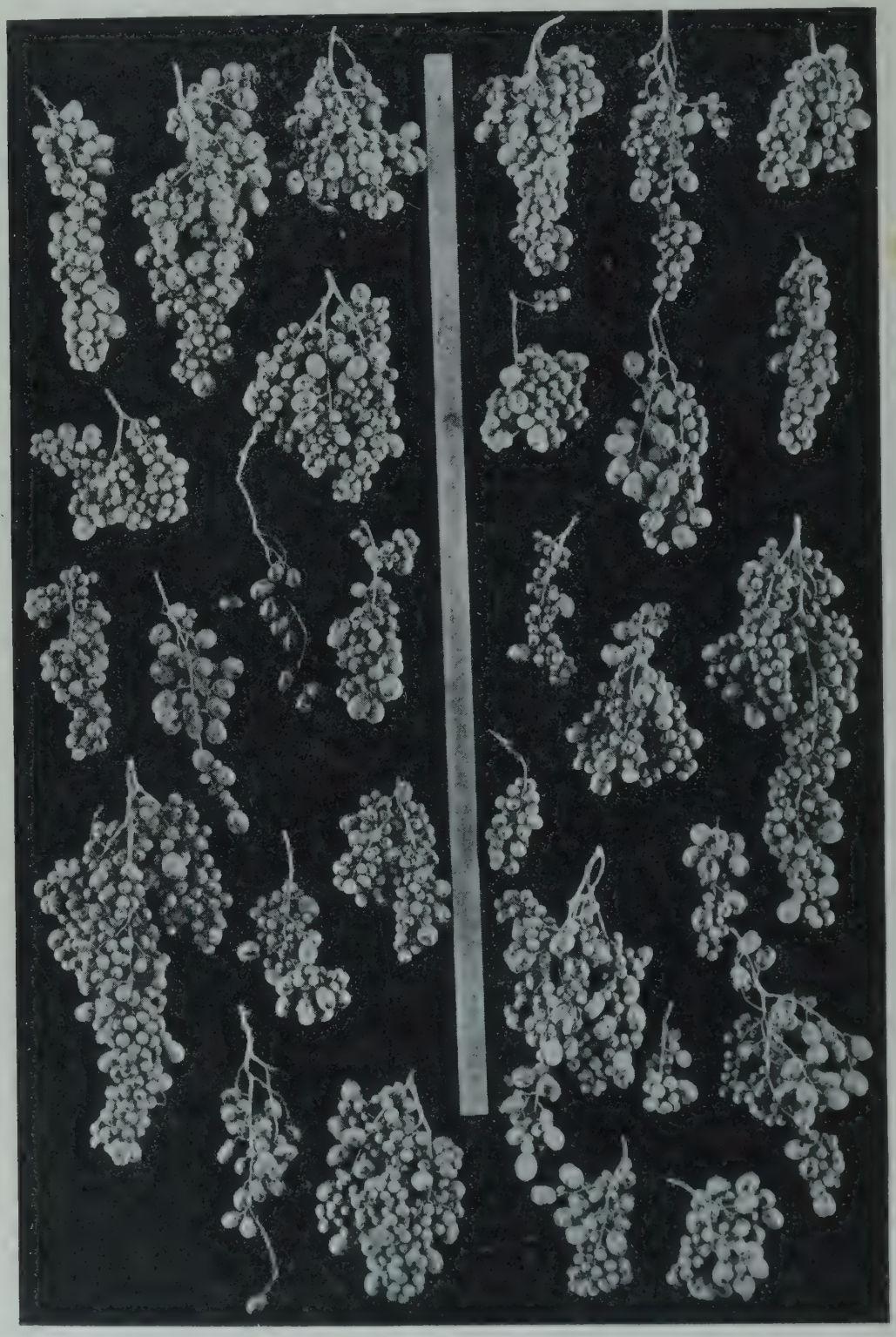

Fig. 3A. The influence of the type of pruning on the set of normal berries on Hunisa. *Normal pruning* (10 per cent of berries normal). (Entire crop from one average vine.)

In the other varieties which set very few small seedless (shot) berries, the increase in the percentage of normal berries has been relatively small. It has ranged from 3.4 to 17 per cent for the *half-*

Fig. 3B. The influence of the type of pruning on the set of normal berries on Hunisa. *Cane pruning, part crop* (82 per cent of berries normal). (Entire crop from one average vine.)

long and *cane pruned, part crop* vines. In these varieties the more *severe pruning* also had little influence on the percentages of normal berries.

TABLE 4

THE INFLUENCE OF PRUNING ON THE PERCENTAGE OF NORMAL BERRIES TO A BUNCH

Variety	Year	Types of pruning—and percentage of normal berries to a bunch					
		Severe	Normal	Half-long, part crop	Cane, part crop	Non-pruned, part crop	Non-pruned, all crop
Muscat of Alexandria	1924	47	68	93	.....	95	78
Muscat of Alexandria	1925	65	69	.....	94	96	93
Hunisa	1925	2.5	10	33	59	.....	.....
Muscat gigas	1925	81	83	97	95	.....	.....
Henab	1925	96	93	97	97	.....	.....
Molinera	1925	77	85	91	92	.....	.....
Malaga	1925	86	86	89	92	.....	.....
Emperor	1925	83	85	91	93	.....	.....
Ohanez	1925	75	88	91	91	.....	.....

DISCUSSION

The great differences in the germinability of pollen and in the set and in the percentage of normal berries as a result of the less severe pruning indicates that the quality of one or both of the flower parts is improved. For the male part of the flower, the improvement is rather conclusively shown by the increase in the percentage of germination of the pollen. The increase in the number and the percentage of normal berries to a bunch indicates that the female part of the flower also is improved. In the Muscat of Alexandria, Muscat gigas, and Molinera, where the pollen germination on the normally pruned vines was 8, 7, and 16 per cent, respectively, an influence on the female part of the flower may be questioned, since the poor quality of pollen alone may have been sufficient to limit the set of normal berries very considerably. The increase in the set of normal berries under the less severe pruning might then, for these varieties, be entirely a result of the great increase in the germinability of pollen. This could hardly be the case, however, in such varieties as Henab, Malaga, and Emperor where the pollen from the *normally pruned* vines gave a germination of 25 per cent or more.

The improvement in the female part of the flowers is further indicated by the results of pollination tests on the *normally pruned* Muscat of Alexandria vines. In these tests twenty bunches on the *normally pruned* vines were dusted each day with pollen of the

non-pruned, part crop vines until all of the calyptera were off. The influence of the pollen of greater germinability on the set of normal berries on the pollinated as compared to the non-pollinated bunches on the *normally pruned* vines and to the non-pollinated bunches on the *cane* and *nonpruned, part crop* vines is shown in table 5.

TABLE 5

THE INFLUENCE OF POLLINATION WITH POLLEN OF THE NON-PRUNED, PART CROP VINES ON THE SET OF NORMAL BERRIES UNDER NORMAL PRUNING AS COMPARED TO THE SET OF NORMAL BERRIES ON THE CANE AND NON-PRUNED PART CROP VINES

Normal berries	Types of pruning and the pollination treatments			
	Normally pruned		Cane pruned, part crop, not pollinated	Non-pruned, part crop, not pollinated
	Not pollinated	Pollinated		
Number to a bunch	34±1.4	68±6.1	115±3.5	140±2.4
Per cent of total berries	69	76	94	96
Increase in number as a result of—				
1. Pollination		100		
2. Less severe pruning			238	312
Increase in percentage as a result of—				
1. Pollination		10		
2. Less severe pruning			36	39

These data show an increase in both the number and percentage of normal berries to a bunch on the pollinated when compared to the non-pollinated bunches on the *normally pruned* vines. This increase as a result of pollination, however, has not been so great as that following less severe pruning.

The question now arises—How do less severe pruning and non-pruning improve the germination of the pollen and the set of berries? Or, on the other hand—Why do *normal* and *severe* pruning tend to reduce the germination of pollen and the set of normal berries? The answer to these questions is, no doubt, bound up with the nutrition of the flower buds from the time of their differentiation until or even after blooming. Hilgard[6] believed that the failure of Muscat of Alexandria to set normal berries was due to a lack of proper soil fertility. The fertilizers which he recommended for the amelioration of the trouble, however, were ineffective. Müller-Thurgau[8] (1883) and Merjanian[7] (1919) state that the poor nourishment of the flowers

is the chief cause of *coulure* and of the setting of shot berries. Müller observed that in cold cloudy weather, which favors the tendency to *coulure,* the elaboration in the leaves and the transport of organic substances to the flowers was limited. This limiting of the food supply to the flowers was especially noticeable when there were a considerable number of rapidly growing shoots on the vine. Both of these workers succeeded in moderating the tendency to *coulure* and *Millerandage* by pinching and ringing. Pinching, according to Bioletti,[3] however, is weakening to such an extent that after one or two years of its practice, production falls off. Sartorius[9] states that the stage of development of the embryonic flowers in the fruit bud at the time growth begins in spring greatly influences their bloom, set, and later development. He[10] has shown also that the poor development of pollen in cold weather may play a considerable role in the dropping of flowers. In deciduous trees it has been stated by Dorsey[4] that the vigorous spurs set in greater number than weak ones, probably because of their larger supply of stored food and because of their greater ability to compete for water and other food substances.

The data of table 6 and figure 4 seem to indicate that the number of leaves on a vine, especially during the early part of the season, as influenced by pruning, may be responsible for the difference in the germinability of its pollen and the set of its berries. For the five varieties these figures show an average increase in the number of leaves to a vine over that for the *normally pruned* vines of 75 per cent for the *half-long pruned, part crop,* and 139 per cent for the *cane pruned, part crop* vines. Then, too, as illustrated by the graphs of figure 4, the *non-pruned* vines had produced more leaves at the time of the first count, on May 15th, two weeks before blossoming, than the *normally* or *severely pruned* vines produced during the entire growing season. The rate of increase in the number of leaves for the remainder of the season was also greater for the *non-pruned* than for the *Normally* or *severely pruned* vines. The graphs of figure 4 show also that the slight decrease in the weight of the individual leaves produced by the *non-pruned* vines was of little importance compared to the great increase in the number of leaves.

The influence of the number of leaves at or near the time of blooming on the germination of pollen and the set of normal berries is further indicated by the ratios shown in table 7.

In view of the data it appears probable that the increase in the number of leaves as a result of less severe or no pruning has resulted in a better nutrition of the flower buds. This better nourishment has

TABLE 6

THE NUMBER OF LEAVES TO A VINE AT OR NEAR THE TIME OF BLOOM UNDER THE DIFFERENT TYPES OF PRUNING

Variety	Beginning of bloom	Full bloom	Time of leaf counts	Types of pruning—and number of leaves			
				Severe	Normal	Half-long, part crop	Cane, part crop
Muscat of Alexandria	May 17	May 25–28	May 5	34	89	.....	470
			June 24	545	773	.....	1724
Muscat gigas	May 13	May 19–23	May 5	101	147	277	489
			June 23	315	436	703	971
Hunisa	May 18	May 25–28	May 7	147	179	517	651
			June 20	891	1134	1437	1897
Ohanez	May 12	May 20–24	May 14	467	476	864	1057
Dizmar	May 13	May 21–25	May 15	258	353	756	1125

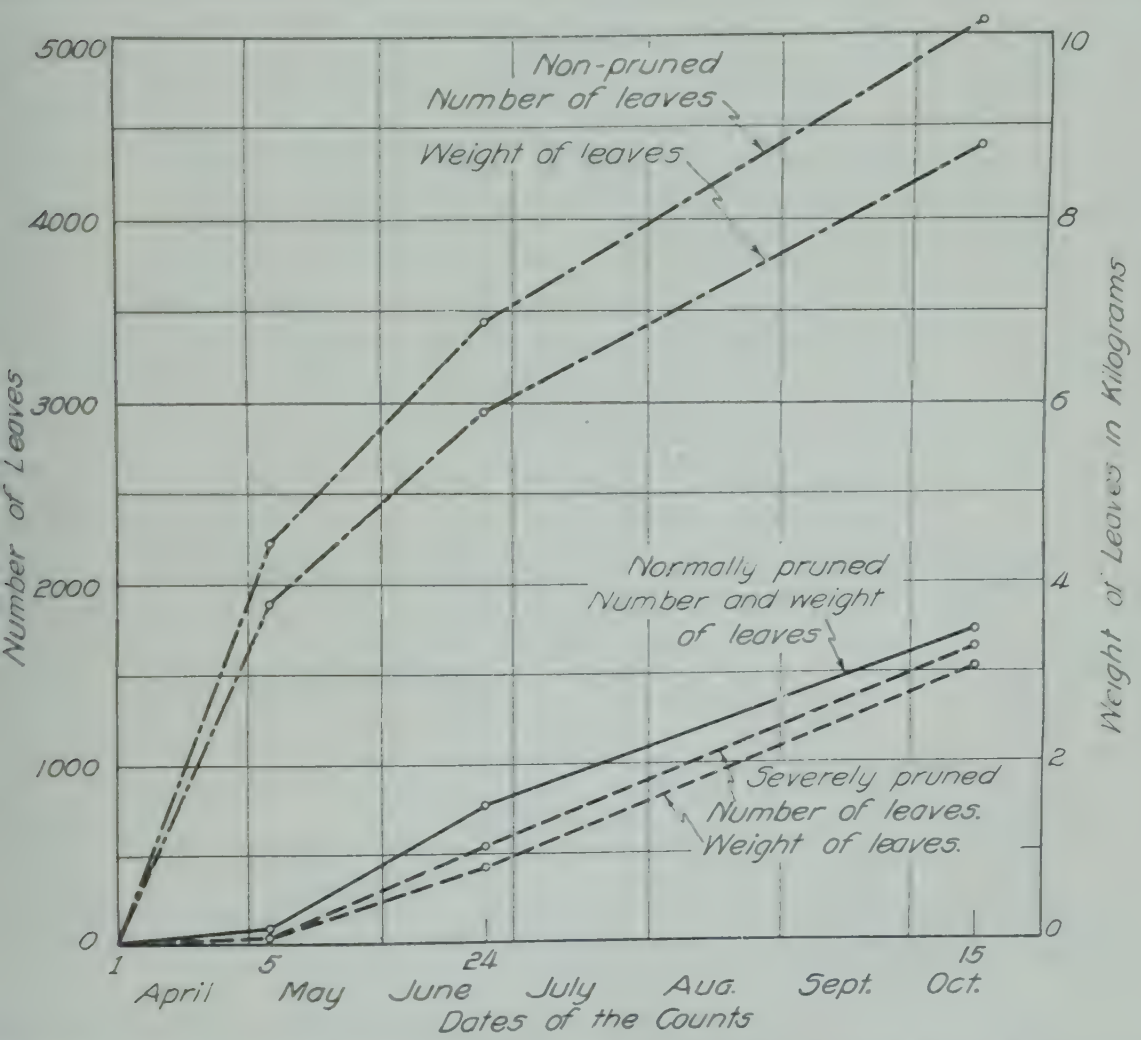

Fig. 4. The number and weight of leaves on a vine at several dates during the season under *severe*, *normal*, and *no pruning*.

given rise to stronger flower parts which, in turn, has resulted in the production of pollen of greater germinability and an increase in the set of normal berries. This relation of the number of leaves to the improvement in set of berries is further substantiated by the fact that the second crop, the bloom for which develops in mid-season when the vine is in full leaf, usually sets normal berries, however prone the variety may be to set shot berries in the primary crop.

TABLE 7

THE RATIOS OF INCREASE IN THE NUMBER OF LEAVES TO THE INCREASE IN THE GERMINATION OF POLLEN AND TO THE INCREASE IN THE SET OF NORMAL BERRIES FOR THE NORMAL, HALF-LONG, AND CANE PRUNED OVER THE SEVERELY PRUNED VINES

Ratios	Normally pruned	Half long-pruned, part crop	Cane pruned, part crop
Leaves/berries	6.5	6.7	7.2
Leaves/germination of pollen	42.8	44.7	42.6

The results of other investigators indicate also that the number of leaves at or near the time of blooming influences the set of fruits. Müller-Thurgau[8] and Sartorius[10] were able to induce *coulure* by the removal of the large leaves. Then, too, if they pinched the tip of the shoot, thus stopping elongation and reducing the keenness of the competition of the flowers for food substances, the tendency to *coulure* was moderated. Sorauer[11] states that "if the wood is thinned too much, i.e., too many leaf branches are cut away in order to furnish light for the blossoms and young fruit, the buds, blossoms and young fruit may be dropped." In work with apples Haller and Magness[5] have found that the number of fruits dropping decreased with an increase in the leaf area per fruit until a certain leaf area was attained.

SUMMARY

Less severe pruning increases germinability of pollen:

When the pollen of the *normally pruned* vines is taken as a standard, the germinability of the pollen of the *half-long pruned, part crop* vines was increased from 38 to 277 per cent, that of the *cane pruned, part crop* vines, from 44 to 606 per cent; that of the *non-pruned, part crop vines*, 219 to 606 per cent; and that of the *non-pruned, all crop* vines, 117 to 576 per cent.

Severe pruning resulted in decreased germinability of the pollen in most of the tests.

Less severe pruning increases set of normal berries:

In Muscat of Alexandria, Muscat gigas, and Hunisa which are very subject to *coulure*, the increase in the set of normal berries to a bunch over that of the *normally pruned* vines was 114, 220, and 407 per cent for the *half-long pruned, part crop* and 238, 320, and 728 for the *cane pruned, part crop* vines, respectively. The increase in Muscat of Alexandria for the *non-pruned, part crop* vines was 266 per cent and for the *non-pruned, all crop* vines was 62 per cent.

In the other varieties—Molinera, Henab, Emperor, Malaga and Ohanez—which are little affected by *coulure*, the increase in the set of normal berries to a bunch ranged from 19 to 112 per cent for the *half-long pruned, part crop* and from 20 to 115 per cent for the *cane pruned, part crop* vines.

The set of normal berries on the *severely pruned* has been less than that on the *normally pruned* vines in six of the eight varieties tested.

Less severe pruning increases percentage of normal berries to a bunch:

In Muscat of Alexandria and Hunisa varieties, which are very subject to *millerandage*, the percentage of normal berries to a bunch was increased 36 and 230 per cent, respectively, on the *half-long pruned, part crop* vines, and 36 and 590 per cent, respectively, on the *cane pruned, part crop* vines. Similar increases were obtained under the types of no pruning with the Muscat of Alexandria.

In the other varieties, which usually set very few small seedless berries, the increase in the percentage of normal berries to a bunch ranged from 3.4 to 17 per cent for the *half-long* and *cane pruned, part crop* vines over that of the *normally pruned* vines.

Severe pruning has reduced the percentage of normal berries below that of the *normally pruned* vines in six of the eight varieties tested.

Flower parts are probably improved under less severe types of pruning:

The increased germinability of pollen, the set of normal berries, and the percentage of normal berries to a bunch, together with the pollination tests, indicate that both the male and female parts of the flowers are improved by the less severe pruning.

The improvement in the flower parts appears to follow as a result of an earlier development of the foliage and an increase in its area with less severe pruning.

LITERATURE CITED

[1] Bradbury, D.
1925. Notes on the dropping of immature sour cherry fruits. Proc. Amer. Soc. Hort. Sci. 22: 105–110.

[2] Bioletti, F. T. and H. E. Jacob
1924. Head, cane and cordon pruning of vines. California Agr. Exp. Sta. Cir. 277: 1–32.

[3] Bioletti, F. T. and F. C. H. Flossfeder
1918. Topping and pinching vines. California Agr. Exp. Sta. Bul. 296: 371–384.

[4] Dorsey, M. J., and H. E. Knowlton
1925. The relation of growth to fruitfulness in some varieties of apple. Proc. Amer. Soc. Hort. Sci. 22: 161–172.

[5] Haller, J. H., and J. R. Magness
1925. The relation of leaf area to the growth and composition of apples. Proc. Amer. Soc. Hort. Sci. 22: 189–196.

[6] Hilgard, E. W.
1884. The Muscat on the southern mesas. California Agr. Exp. Sta. Bul. 17: 1.

[7] Merjanian, A. S.
1919. De la coulure et du millerandage (Russian with French summary). Bul. Sta. Oenolog., Odessa, 1: 1–55.

[8] Müller–Thurgau, H.
1883. Über das abfallen der Rebenblüten und die entstehung kernloser Traubenbeeren. Der Weinbau no. 22.

[9] Sartorius, Otto
1926. Zur Entwicklung und Physiologie der Rebblüte. Angew. Botanik 8: 29–62.

[10] Sartorius, Otto
1926. Zur Entwicklung und Physiologie der Rebblüte. Angew. Botanik 8: 65–89.

[11] Scrauer, Paul
1924. Handbuch der Pflanzenkrankheiten. 5th Ed. 1: 396–398.

[12] Winkler, A. J.
1926. Some responses of Vitis vinifera to pruning. Hilgardia 1: 526–543.

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SOME FACTORS AFFECTING THE IRRIGATION REQUIREMENTS OF DECIDUOUS ORCHARDS

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(Contribution from the Division of Irrigation Investigations and Practice coöperating with the Division of Pomology, College of Agriculture, University of California, and with the Division of Agricultural Engineering, Bureau of Public Roads, United States Department of Agriculture, and the Division of Engineering and Irrigation, California State Department of Public Works.)

INTRODUCTION

The relation of water to plant growth is of especial interest to growers of deciduous fruits. Consideration of the relative losses of moisture from irrigated soils by evaporation or transpiration through plants and by means of surface evaporation from the soil is becoming increasingly important. This is especially true in many of the deciduous fruit areas of California, where the cost of irrigation is one of the main items of expense in orchard management. The orchardist growing fruit in an arid or semi-arid region, where irrigation is necessary, supposedly has an advantage over the grower of similar fruit in a humid area, because the supply of moisture in the soil can be controlled to a greater extent. Therefore, it is important to consider the effect of different degrees of soil moisture on the use of water by trees, and the effect of such differences on the kind of fruit produced.

A portion of the California deciduous fruit orchards have been planted in localities where dependence has been placed upon rainfall only. However that irrigation is necessary in many sections of California for the best production of horticultural crops is a fact now coming into recognition. In many of these areas, previously unirrigated, gravity water is not available and recourse must be had to

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pumping from underground supplies. The orchard land awaiting development in California is generally the higher land of the valleys and the adjacent foothill areas. Here, as well as in the pumping areas, water for irrigation is relatively limited. Proper use of the available supply is therefore of vital importance. The older irrigated fruit sections are also confronted with many problems in the use of water. In these sections, the too lavish use of water has probably been the contributing cause of many troubles.

Because of the wide variations in soils in the different deciduous fruit areas of the State and of the extreme differences in topography, irrigation methods must, and do, vary widely. In the citrus groves of the State, irrigation practice has followed certain arbitrary rules. Deciduous orchard irrigation practice, on the other hand, has no semblance of uniformity. Very little definite information is available concerning the efficacy of different practices, especially as to the frequency of irrigation and the amount of water to apply. However, it is not the purpose of the present report to deal directly with methods or practices in deciduous orchard irrigation, but to discuss the results of certain experiments, which, it is believed, will afford a better understanding of their problems.

The experiments fall into four classes and will be discussed in the following order: first, observational data obtained from commercial orchards in the Santa Clara Valley, together with records of moisture conditions in these orchards for a period of four years; second, data obtained from a small block of trees at the Branch of the College of Agriculture, University of California, at Davis; third, studies of the behavior of young trees in potometers or tanks under controlled conditions; and fourth, a comparison of the losses of moisture by evaporation and through transpiration from the same soils, and studies of losses of moisture by evaporation from the soil in tanks and from field plots. The latter studies were supplemented by some experiments on the movement of soil moisture.

Briefly stated then, this report has for its purpose the presentation of certain data relating to soil moisture conditions prevalent in commercial orchards, with an attempt to analyze causes resulting in losses of moisture from irrigated soils.

Studies covering water relations of plants, the movement of moisture through the soil, the losses of moisture from soils, and the control of soil moisture by tillage, probably constitute the bulk of agricultural literature. They are so numerous that only brief mention of pertinent matter can be made in the order of the discussion in this report.

SECTION I

IRRIGATION STUDIES IN SANTA CLARA VALLEY PRUNE ORCHARDS

The Santa Clara Valley of California is one of the oldest and best established deciduous fruit areas of the State. The principal fruit crop is prunes, and for this reason, prune orchards were selected for study. Early in the spring of 1919, a system of soil sampling was begun in a number of mature prune orchards in this valley, in order to study the moisture behavior in response to different irrigation practices.

At present, practically all orchards for which a water supply is available are irrigated. The census of 1910 reports Santa Clara County with an irrigated acreage of 37,637, and the 1920 census reports 71,274 acres, an increase of 89.4 per cent. The mean annual rainfall at San Jose, as computed in 1922, is 16.79 inches, an amount sufficient, on certain types of soil, to produce profitable crops of fruit without irrigation. The fact that trees will not always be permanently injured if irrigation is withheld affords excellent opportunity to study the behavior of trees and the variation of soil moisture through long periods during the growing season, without the necessity of replenishing the moisture supply in order to keep the trees alive.

The rainfall during each of the four years, 1919 to 1922, during which the Santa Clara Valley orchards were under observation, was less than normal. The precipitation at San Jose during 1919 was 11.98 inches. That of 1920 was 8.5 inches; of 1921, 14.59 inches; and of 1922, 16.49 inches. Most of the rain fell during the period from November to March. The amount during the growing season was insignificant. At times during the summer the temperature exceeded 90 degrees Fahrenheit and occasionally temperatures higher than 100 degrees were recorded. Complete climatological records taken at San Jose can be obtained from the annual summaries published by the United States Weather Bureau.

Orchards Selected for Observation

A careful canvass of the valley was made and certain mature prune orchards were selected for observation. These orchards, which are comparable as to age and condition, are all located in the central portion of the valley, between San Jose and Saratoga, within about two miles of each other. They are all on a type of soil classed in the soil survey as Yolo clay loam, and locally known as sediment soil, which is considered best for fruit. The orchards which have been selected from among those under observation, and for which data are presented in this bulletin, are listed in table 1. The yields of fruit from some of these orchards had to be ascertained from larger acreages than were intensively sampled to obtain the records of the moisture conditions. In every case, however, the larger acreage received the same irrigation treatment as that in which intensive sampling was done. Sampling was restricted to the smaller acreages in each case because of the difficulty of adequately sampling the larger areas.

The number of producing, young non-bearing, and inferior trees, the blank spaces, and odd varieties are listed in table 1. This was the condition of each orchard at the end of the growing season of 1919.

TABLE 1

Santa Clara Valley Prune Orchards and the Condition of Trees at the End of the Growing Season, 1919

Orchard	Root stock	Date of planting	Acreage intensively sampled for moisture determinations	Acreage from which fruit yields were obtained	Good producing prune trees	Inferior mature prune trees	Young non-bearing prune trees	Vacant spaces in orchard	Trees other than prune trees
1	Almond	1895	15.5	15.5	1251	113	132	4	48
2	Peach	1892	9	14	924	5	27	0	27
3	Almond	1895	10	10	610	90	10	20	10
4	Myrobalan	1893	9	19	645	2	14	1	14
5	Almond	1898	10	10	658	20	41	3	24
6*	Almond	1896	21	25	1804	74	63	68	14

* 80 trees were removed from this orchard at the end of the growing season, 1919.

Orchard No. 1 was selected as a typical unirrigated orchard in the Santa Clara Valley. It had not been irrigated since 1912, and before this time had received only light applications of water at infrequent intervals. The condition of the orchard, as indicated by the number of replants, vacant places, and inferior trees, is indicative of injury which may be partly attributed to drought.

Orchard No. 2 was selected as an example of a winter irrigated orchard. The irrigation treatment in this orchard has not varied for many years. It has always been irrigated during the dormant season if water was available, but never during the summer.

At the time the observations started, orchard No. 3 was essentially a non-irrigated orchard. Water was applied once in 1917, for the first time in seven years, but not in 1918. It was understood when observations were started that orchards No. 1 and No. 3 were not to be irrigated. The owners of these orchards declared they did not believe irrigation was beneficial. However, as will be seen, these orchards were irrigated after the season of 1919.

Orchards No. 4 and No. 5, which are adjacent, were selected as typical irrigated orchards. Orchard No. 4, previous to 1919, usually received one summer irrigation. Subsequently, however, it received two or three applications of water each year. Orchard No. 5, before 1919, had been irrigated at least twice each summer, which was substantially the same practice during the following four years.

Orchard No. 6, up to the year 1917, had been irrigated during the dormant season with gravity water. After the season of 1917, the orchard was summer-irrigated and when it was selected in 1919 it was thought to be a well-irrigated orchard.

Cultural Methods in the Orchards

There were no essential differences in the handling of the trees in the different orchards, with the exception of cultivation and irrigation. Orchard No. 4 was pruned more heavily in the winter of 1920 than any of the other orchards, but in the other years the amount of pruning was substantially the same in all of the orchards. The spray treatments for brown apricot scale and red spider were likewise about the same in each orchard. However, it was observed that orchard No. 5 apparently had more brown apricot scale in the fall of 1922, than any of the others.

On five acres of orchard No. 4, 26 tons of chicken manure were applied in October, 1920. This orchard also received an application of 2 tons of sugar beet lime to the acre during January, 1921. Forty

trees in orchard No. 6 received an application or sodium nitrate in February, 1920. No other fertilizer was added during the period of observation.

Cover crops were planted in the fall of each year in each orchard. Sweet clover (*Melilotus indica*) was usually planted, and if the fall irrigation was late, barley was planted with it. In some orchards, owing to the impracticability of planting sweet clover early enough, vetch was used in its place. There were no appreciable differences in the dates of plowing and disking under of the cover crops in the spring of each year. The dates of plowing and disking in the spring for the four years did not materially vary, the earliest date of plowing having been March 25 and the latest date, April 18.

The management of the soil varied markedly in the different orchards. Orchard No. 1 was plowed, disked and harrowed in the spring of 1919. It received one cultivation, and the soil was smoothed down before the prunes dropped. In 1920, the orchard was not plowed, but was disked and harrowed just after the irrigation in April, and received one cultivation and smoothing in June. It was disked and harrowed again after the irrigation in October, 1920. It was plowed in April, 1921; and after the irrigation in May, it was disked and harrowed. This orchard received little cultivation other than that necessary to destroy the cover crop, and to prepare the land before and after irrigation.

Orchard No. 2 was the most frequently cultivated of any orchards observed in the Santa Clara Valley. This practice had not varied for many years. After plowing in the spring, the orchard was cultivated at intervals, usually not exceeding 10 days, until the props were placed under the trees and the prunes began to fall. Immediately after the crop was removed, cultivations were again started and continued until the cover crops were planted. The condition of the soil and of the trees is illustrated in figure 1. This photograph was taken on October 4, 1920, and may be compared with figure 2, which was taken in orchard No. 5 on November 1, 1920. The excellent condition of the trees in orchard No. 2, as indicated by the scarcity of replants and inferior trees and the absence of vacant spaces, is proof of the extreme care the owner has bestowed upon them in every way including cultivation. There were no vacant spaces and only five inferior trees in the 14 acres comprising orchard No. 2. The odd varieties were planted around the dwelling, and the replants were made necessary by the construction of a railroad along one side of the orchard.

Fig. 1. Orchard No. 2 on October 4, 1920, showing condition of soil and trees.

Fig. 2. Orchard No. 5 on November 1, 1920.

Orchard No. 3 was poorly cared for up to 1920. The poor condition of the trees indicated the lack of attention for four or five preceding years. This and orchard No. 1 were selected as examples of non-irrigated orchards. They had not been irrigated for a number of years. They were typical of non-irrigated orchards in this location, and showed the effect of drought. Orchard No. 3 was not properly plowed in the spring of 1919. The strips between the trees in the direction of plowing were not disturbed. Later in the season of 1919, an attempt was made to cultivate the orchard and destroy the weeds growing in these areas, but the soil was too dry to be disked. During 1920, 1921, and 1922, the management of orchard No. 3 was entirely changed. The orchard was thoroughly plowed each spring and was cultivated at intervals of a week or ten days until the prunes began to drop.

Orchards Nos. 4, 5, and 6 were given practically the same soil management. The practice was to plow in the spring, usually early in April, and then double disk both ways. In one or two instances the soil was harrowed with a spike-tooth harrow. Before irrigation, levees which formed the basins around each tree were thrown up with a disk, and a supply ditch was constructed. Following irrigation, the levees were leveled with a disk, and the soil disked both ways. Whenever the irrigation was the last one before the crop ripened, the soil was smoothed off with a drag to receive the falling prunes. These orchards were not cultivated if there were no weeds.

Irrigation of the Orchards

The total amounts of water applied by irrigation, the size of the stream used in irrigation, the total number of hours, the average depth of penetration of the water applied, and the cost of the water is given in table 2.

Orchard No. 2 was irrigated with gravity water, while all the others were irrigated with water pumped from wells. In every case, the depth of water in the wells was in excess of 100 feet, the average pumping lift being about 135 feet. The cost of water under this condition is necessarily high. On some of the other orchards which were under observation, but which are not listed here, the costs for water were even higher than those recorded in table 2. In one instance, water which cost $103.85 an acre foot, was purchased for the irrigation of 25 acres. It is obvious, therefore, that economical use of water is of great importance in this locality.

TABLE 2

AMOUNT OF IRRIGATION WATER APPLIED TO THE SANTA CLARA PRUNE ORCHARDS, WITH COSTS OF THE WATER AND AVERAGE DEPTHS OF PENETRATION

ORCHARD NO. 1

Year	Dates of irrigation		Hours to the acre	Size of stream, gallons per minute	Depth of water applied in inches per acre	Average depth of penetration in feet	Cost of water for each acre in dollars	Cost of water for each acre-foot in dollars
	Begun	Finished						
1920	Apr. 17	Apr. 24	7.55	225	4.01	5.0	11.32	33.86
	Sept. 28	Oct. 2	3.42	455	3.45	2.0	8.55	29.70
1921	May 6	May 11	6.32	476	6.62	10.5	15.80	28.55
	Nov. 11	Nov. 14	4.65	455	4.65	4.5	11.62	29.95

ORCHARD NO. 2

1919	Feb. 28	Mar. 2	2.66	2000	12.00	12.0	2.22	2.22
1920	Mar. 22	Mar. 24	2.66	2000	12.00	9.0	2.22	2.22
1921	Feb. 19	Feb. 21	8.00	1500	14.15	10.0	6.66	5.66
1922	Feb. 14	Feb. 16	2.66	2000	12.00	12.0	2.22	2.22

ORCHARD NO. 3

1919	Nov. 23	Dec. 2	10.70	156	3.68	.....	16.05	52.28
1920	Apr. 30	May 5	10.00	130	2.88	6.0	15.00	62.50
	June 20	June 23	3.50	468	3.62	6.5	7.88	26.07
	Oct. 20	Oct. 24	4.25	431	4.50	6.5	8.50	22.65
1921	May 21	May 24	8.70	314	6.04	9.5	13.05	25.95
	Oct. 14	Oct. 17	6.00	304	4.02	3.0	9.00	26.87
1922	May 15	May 18	9.50	306	6.36	10.5	14.35	25.19
	Oct. 16	Oct. 20	7.20	320	5.10	4.5	10.80	25.39

ORCHARD NO. 4

1919	May 27	June 3	15.75	242	7.73	9.0	23.63	36.70
	Nov. 21	Nov. 29	8.22	314	5.70	.....	12.33	25.93
1920	May 20	May 24	11.44	200	5.08	.....	17.16	36.50
	June 15	June 19	5.11	432	4.88	12.0	10.22	22.60
	Nov. 1	Nov. 6	5.11	431	4.87	5.0	10.22	22.60
1921	May 20	May 24	5.55	391	4.79	12.0	12.50	28.19
	Oct. 6	Oct. 9	5.55	337	4.13	2.5	8.33	21.80
1922	Apr. 26	May 2	10.00	321	7.06	12.0	15.00	25.52
	June 6	June 12	7.78	323	5.56	12.0	11.66	22.70
	Oct. 10	Oct. 14	7.22	315	5.00	3.0	10.83	25.60

TABLE 2—(*Continued*)

ORCHARD NO. 5

Year	Dates of irrigation		Hours to the acre	Size of stream, gallons per minute	Depth of water applied in inches per acre	Average depth of penetration in feet	Cost of water for each acre in dollars	Cost of water for each acre-foot in dollars
	Begun	Finished						
1919	June 9	June 15	18.6	165	6.80	12.0	27.90	49.46
1920	Mar. 19	Mar. 25	14.65	341	11.10	9.0	29.30	31.68
	Sept. 18	Sept. 20	4.70	431	5.00	4.0	9.40	22.53
1921	May 20	June 2	7.00	391	6.01	8.0	15.75	31.50
	Sept. 25	Sept. 28	6.95	425	6.52	3.0	15.64	28.79
1922	May 29	June 5	8.00	444	7.82	11.0	16.00	24.53
	Oct. 11	Oct. 16	9.00	372	7.38	4.5	18.00	29.25

ORCHARD NO. 6

1919	May 2	May 11	8.38	320	5.92	6.0	12.57	25.57
	June 13	June 18	6.97	303	4.34	3.5	9.84	26.85
	Oct. 4	Oct. 14	6.28	303	4.20	3.5	9.43	26.85
1920	Feb. 14	Feb. 23	7.42	341	4.97	4.5	11.14	26.90
	May 6	May 15	10.00	223	4.99	6.0	14.99	36.09
	Sept. 20	Sept. 25	5.43	355	4.25	3.5	9.50	26.82
1921	Apr. 29	May 6	6.28	373	5.17	6.0	11.00	25.55
	Sept. 30	Oct. 6	8.05	320	5.58	3.5	14.08	30.40
1922	May 29	June 17	9.38	321	6.62	9.0	16.89	30.40
	Sept. 25	Oct. 9	7.76	337	5.77	4.5	13.58	28.29

All irrigation was applied by the basin method. The same amount of water was given to each tree, insuring a very uniform distribution of moisture in the soil. The evenness of the application is illustrated in figure 3, which was taken during the irrigation of orchard No. 4, from November 1 to November 6, 1920. The water applied was measured by means of weirs placed in the main delivery ditches, or at the outlets of concrete pipe delivery stands.

YIELDS OF FRUIT FROM THE SANTA CLARA ORCHARDS

The yields of fruit from the orchards are reported in table 3. The fresh and dried fruit yields, the pounds of fresh fruit required to make one pound of dried fruit, the average size of dried prunes,

which were calculated from the weight of fruit as graded at the packing house, the yield to the acre, and the yield to the tree are given. The acreage upon which the yields to the acre are based is the total acres in the orchard. The number and the condition of the trees were noted each year. The record of the condition of the orchards at the end of the 1919 season is given in table 1. The inferior trees were carefully noted, and in the final count of the number of trees upon which to base the yield to the tree, these were

Fig. 3. Irrigation of orchard No. 4, November 1, 1920. The same amount of water is applied in basins around each tree, insuring a uniform distribution of moisture.

grouped so that a certain number of inferior trees were taken to be equivalent to one good tree. The estimated yield to the tree, or the yield to 75 trees, the usual number to the acre when planted 24 feet apart, probably will afford a better basis for comparison than the yield to the acre. The trees in orchard No. 1 were planted 20 feet apart. In all of the other orchards the trees were spaced 24 feet apart. The fruit from orchard No. 1, for the years 1921 and 1922, was mixed with that from another orchard. Therefore the yields are not recorded for these years.

TABLE 3

YIELDS OF FRUIT, FROM THE SANTA CLARA VALLEY PRUNE ORCHARDS, FOR 1919, 1920, 1921, AND 1922

Orchard	Acres in orchard	Fresh fruit, pounds	Dried fruit, pounds	Pounds of fresh fruit to make one pound of dried fruit	Average size in number of dried fruit to the pound	Yield to the acre		Yield to the tree		Yield to 75 trees	
						Fresh fruit, pounds	Dried fruit, pounds	Fresh fruit, pounds	Dried fruit, pounds	Fresh fruit, pounds	Dried fruit, pounds
Yields, 1919											
1	15.5	191,474	96,377	1.99	79	12,353	6,214	150	75.5	11,250	5,662
2	14.0	205,281	97,168	2.11	86	14,663	6,941	221	104.8	16,575	7,860
3	10.0	60,000	35,534	1.69	73	6,000	3,553	95	56.4	7,125	4,230
4	19.0	233,000	115,301	2.02	80	12,363	6,068	195	84.5	14,625	6,337
5	10.0	184,390	61,933	2.98	99	18,439	6,193	276	92.7	20,700	6,953
6	25.0	502,250	200,778	2.50	90	20,090	8,031	273	109.1	20,475	8,182
Yields, 1920											
1	15.5	75,000	34,550	2.17	71	4,837	2,229	59	27.3	4,425	2,047
2	14.0	155,151	68,283	2.27	68	11,082	4,877	168	73.8	12,600	5,535
3	10.0	66,568	27,674	2.41	60	6,657	2,768	105	43.9	7,875	3,293
4	19.0	140,000	65,198	2.15	56	7,369	3,431	98	48.2	7,350	3,615
5	10.0	62,000	27,428	2.26	56	6,200	2,743	94	41.5	7,050	3,113
6	25.0	267,800	107,638	2.49	60	10,712	4,306	150	60.3	11,250	4,522
Yields, 1921											
2	14.0	164,990	74,119	2.23	68	11,785	5,294	179	80.2	13,425	6,015
3	10.0	96,000	42,000	2.29	70	9,600	4,200	152	66.6	11,400	4,995
4	19.0	137,450	67,256	2.04	48	7,234	3,539	95	50.2	7,125	3,765
5	10.0	138,000	58,080	2.38	61	13,800	5,808	212	89.1	15,900	6,683
6	25.0	329,020	144,895	2.27	56	13,160	5,796	184	81.2	13,800	6,090
Yields, 1922											
2	14.0	86,695	44,473	1.95	48	6,193	3,177	94	48.2	7,050	3,615
3	10.0	92,000	24,280	2.64	.....	9,200	2,428	146	38.5	10,950	2,888
4	19.0	123,344	63,516	1.94	45	6,492	3,429	111	49.1	8,325	3,682
5	10.0	116,000	49,020	2.37	59	11,600	4,902	180	76.1	13,500	5,707
6	25.0	206,721	99,785	2.04	49	8,269	3,991	116	55.9	8,700	4,192

Analyses of Fresh Fruit from the Santa Clara Orchards

Samples to determine the sugar content of the fresh prunes were taken from each orchard. Since the number of samples and the amount of fruit which could be analyzed were limited, only one sample, of about five pounds of prunes from each orchard, was selected each year. The fruits comprising the sample were picked up in a systematic manner in each orchard. The locations from which the fruits were taken were numerous enough to be representative of the entire orchard. Sound fruits only were selected, and these were picked up from beneath normally producing trees. The prunes were forwarded to the laboratory in wooden cartons and immediately placed in freezing storage until analysis was begun. In some cases analyses were made immediately; in others, the fruit was placed in cans, sealed, and sterilized, the analytical work being done at a later date. The analyses* of the fresh prunes from the different orchards for each of the four years are given in table 4.

The value is, in each case, the average of duplicate portions which agreed closely. The percentages of sugar listed in the table are the percentages of total sugar in the flesh as invert sugar after inversion.

The results obtained from the analyses of fruits for the season of 1919, 1920, and 1921 seem to be fairly uniform. Prunes from orchard No. 5, collected in 1919, seemed to have a greater percentage of sugar in the flesh, calculated on a dry or water-free basis, than prunes from the other orchards. The sugar content of the prunes of 1921, from orchard No. 2, seemed to be low. The 1922 analyses show greater differences in the sugar content.

These differences may be due to unavoidable errors in sampling. Not enough samples were analyzed to determine the variability due to this cause. The difficulty of selecting a representative sample in the field may be so great that errors are introduced. Denny[25] has shown the variation in results obtained in the analyses of 51 fruits taken from one tree, and his work suggests that large numbers of samples must be taken to give assurance that the differences observed are not due to random sampling.

However, it is clear that the differences found in these analyses can not be attributed entirely to the effect of irrigation. The sugar content of the fruit from orchards with low soil moisture during the growing season, in which are included orchards Nos. 1, 2, and 3,

* The analyses of the samples for the years 1919, 1920 and 1921 were made by Prof. A. W. Christie, and the 1922 analyses were made by Mr. H. Goss, of the University of California.

averaged 70.4 per cent for 1919, 65.6 per cent for 1920, and 66.0 per cent for 1921; while orchards 4, 5, and 6, in which the soil-moisture supply usually was greater, averaged 72.3 per cent, 66.0 per cent, and 67.0 per cent for these years. The differences in these values are no greater than those Denny[25] suggests might be due to random sampling.

TABLE 4

ANALYSES OF FRESH PRUNES FROM THE SANTA CLARA ORCHARDS

Orchard No.	Dates samples collected	Pit, percentage of total weight	Flesh, percentage of total weight	Water in flesh	Sugar in flesh	Sugar in flesh on dry basis
				Per cent	*Per cent*	*Per cent*
1	Sept. 2, 1919.....	9.4	90.6	67.6	23.0	71.0
2	Sept. 2, 1919.....	9.9	90.1	68.7	21.8	69.3
3	Sept. 2, 1919.....	9.4	90.6	66.8	23.5	70.8
4	Sept. 2, 1919.....	9.1	90.9	69.6	21.8	71.7
5	Sept. 2, 1919.....	8.5	91.5	71.1	21.9	75.8
6	Sept. 2, 1919.....	6.9	93.1	70.3	20.9	70.4
1	Sept. 10, 1920...	12.5	87.5	63.7	23.4	64.5
2	Sept. 10, 1920...	9.6	90.4	65.0	23.0	65.7
3	Sept. 10, 1920....	12.0	88.0	64.4	23.7	66.6
4	Sept. 10, 1920....	12.3	87.7	66.1	22.2	65.5
5	Sept. 10, 1920....	12.8	87.2	65.4	22.4	64.7
6	Sept. 10, 1920....	12.3	87.7	63.6	24.7	67.9
1	Sept. 8, 1921.....	9.9	90.1	66.45	23.05	68.7
2	Sept. 8, 1921.....	10.8	89.2	64.5	22.4	62.95
3	Sept. 8, 1921.....	11.0	89.0	66.1	22.5	66.25
4	Sept. 8, 1921.....	10.9	89.1	63.95	25.2	69.9
5	Sept. 8, 1921.....	9.9	90.1	66.7	22.2	66.6
6	Sept. 8, 1921.....	11.2	88.8	66.0	22.8	66.9
2	Sept. 10, 1922....	5.0	95.0	65.9	17.8	52.2
3	Sept. 10, 1922....	4.5	95.5	69.1	15.6	50.5
4	Sept. 10, 1922....	4.5	95.5	64.9	20.9	59.6
5	Sept. 10, 1922....	4.3	95.7	73.3	18.6	69.7
6	Sept. 10, 1922....	4.8	95.2	71.0	17.2	59.3

There appears to be no consistency in the relation of irrigation to sugar content. When the percentages of sugar found are compared with the soil-moisture history of the corresponding orchards, it will be seen that in some years the orchards which had the greatest amount of moisture in the soil had the most sugar in the fruit; but in other years, orchards with less moisture in the soil had the most sugar in the fruit.

Soil-Moisture Conditions in the Santa Clara Orchards

Before the routine system of soil sampling was inaugurated, a thorough soil survey was made of the orchards. Places where variations in the upper six feet of soil occurred were recorded. The places where the samples were to be taken were then decided upon in relation to these variations in soil in order that they might be representative. At least six places were selected for sampling in each orchard.

Since the variations in the results obtained in sampling orchard soils for moisture content are so great, it is believed that samples taken at random in an orchard will not give comparable results, even though a large number be taken. It was found that comparable results could be obtained, however, if the samplings were confined to definite places in the orchards, such locations being representative of the entire area under observation. In all of the soil-moisture sampling reported herein, the samples were taken from the same places throughout the period of observation. The successive samples were not more than 6 or 8 inches from the previous ones at each location, and at the end of four years the final samples were not more than 3 or 4 feet from the place the first samples were taken. Since the application of water in all of the orchards was very uniform, little variation was found because of irregularities in the wetting of the soil.

Trials with devices commonly used to take soil samples showed that the soil tube was the most satisfactory tool to use. Comparative tests between augers and the soil tube indicated that with the use of the latter, maximum amounts of moisture were found in the samples. These trials also indicated that comparable results could be obtained only when all of the soil removed from the hole was used in making the moisture determination. Subdividing the sample usually resulted in inaccuracies. For these reasons all samples from the Santa Clara Valley orchards, and in fact all those used in these investigations after 1918, were taken with the soil tube. This tube had a drive point so shaped that the core of soil could be cut without compacting either the core itself or the soil ahead of the cutting point.

Soil samples for moisture determinations were taken before and after each irrigation and usually at intervals of from two to three weeks during the summer months.

A further source of error in making moisture determinations was found in the incompleteness of the drying of the samples. There was

a variation from day to day in weights of soil which supposedly had been thoroughly dried, but which were held in the oven with the expectation of reducing them to a constant weight. In all of the work reported herein, samples were repeatedly check-weighed until it was thought a minimum weight in drying had been obtained. All samples were dried in an electric oven which was regulated to operate between 105 and 110 degrees Centigrade.

Moisture equivalent determinations were made on many of the samples. It was intended to record the moisture percentages as ratios of the moisture equivalent, or of any of its related soil-moisture constants. However, wide variations were found in the moisture equivalent values when repeated determinations were made on the same sample of soil. Some of the causes of these variations have since been discussed by Veihmeyer, Israelsen, and Conrad.[51] These authors found that the value for the moisture equivalent is influenced markedly by the amount of soil used in making the determination. Many of the earlier moisture equivalent determinations, in which the weight of soil used in making the determination varied appreciably from 30 grams, are not sufficiently accurate. A further variation, which was sufficient to affect the results when precision was desired, was noted in the value for the moisture equivalent when samples of the same soil were centrifuged at different times, although other conditions were apparently the same. Therefore, too strict an application of the moisture equivalent to the interpretation of soil-moisture data can not be made until the technique of the method is more thoroughly refined. Furthermore, Puri[43] shows that the hygroscopic coefficient can not be satisfactorily determined even when the method is refined beyond that reasonably possible in routine determinations. It appears, then, that the hygroscopic coefficient can not be used as a satisfactory basis for the measurement of the available water in the soil. However, it must be mentioned that, in spite of the variations in moisture equivalent determinations and consequently in the wilting coefficients which are calculated from them, there was a fairly close agreement between the calculated value and the moisture content of the soil at the time the trees so wilted that they did not recover until water was applied to the soil.

The moisture equivalents listed in the following tables are the averages of numbers of determinations made on 30-gram samples of soils, which were taken from the several orchards at different times. The wilting coefficients and hygroscopic coefficients have been calculated from these moisture equivalent determinations.

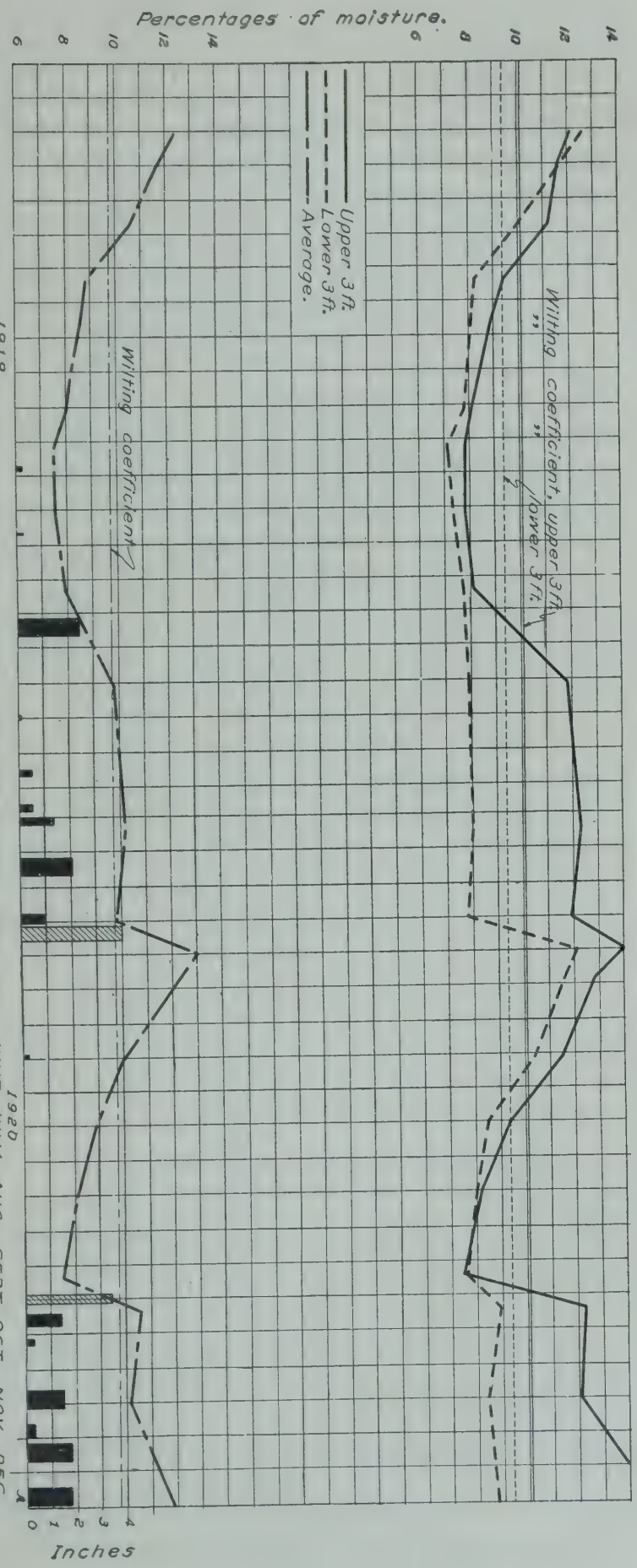

Percentages of moisture.
Upper 3 ft.
Lower 3 ft.
Average.
Wilting coefficient, upper 3 ft.
" " lower 3 ft.
Wilting coefficient
1919
1920
APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC.
Inches

Fig. 4. Soil-moisture conditions in Santa Clara Valley prune orchard No. 1 during 1919 and 1920. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

The percentages of moisture found in the six orchards at different times are graphically illustrated in figures 4 to 15. The average moisture content of the first 3 feet of soil and that of the second 3 feet are shown together with the average moisture content of the first 6 feet. The depth of water in inches applied by irrigation is shown in each case. The heights of the hatched rectangles indicate the total amount of water applied, and the widths of the rectangles, the time it took to apply the water. Rainfall is shown by the solid black rectangles. These show the total rainfall for each month and are placed in a position to indicate the principal dates on which rain fell during the month.

Figures 4 and 5 show that for the years 1919, 1920, and 1921, the soil moisture of the upper 6 feet of soil in orchard No. 1 was reduced below the calculated wilting coefficient for at least two months during the growing season. During the season of 1919, the soil-moisture supply in this depth of soil was reduced below the wilting coefficient about the first of July, and remained dry throughout the remainder of the season. There was no water taken out of the first 6 feet of soil after the middle of September. Apparently the limit of available moisture had been reached by this time. The graph representing the moisture content of the soil to a depth of from 0 to 3 feet is practically parallel with that of the soil from 3 to 6 feet during the summer months of 1919. The soil in this orchard was noted to be very dry at the sampling of July 7, and at all of the subsequent samplings the soil was so dry that great difficulty was encountered in driving the tube into the soil. The trees were decidedly wilted on July 24, 1919. By September 1, it was estimated that about one-half of the leaves of the trees had dropped. The trees were entirely defoliated by November 1. It is a surprising fact that in spite of the low moisture content of the soil to a depth of 6 feet, the trees produced a crop of over 6 tons of green fruit and over 3 tons of dried fruit to the acre. The trees undoubtedly suffered and clearly showed the evidence of lack of water during the latter part of the growing season. When it is remembered that this orchard had undergone this same treatment for many years, the yield is still more surprising. It is true, however, that the yield for 1919 was higher than that usually obtained from this orchard. The average yield up to 1919 had been 3½ tons of green fruit to the acre.

The winter of 1919–1920 was exceptionally dry and the irrigation in April, 1920, of 4.01 acre-inches to the acre, brought the moisture content of the soil up to a point which probably would correspond,

at that time of year, to an amount equal to that usually resulting from rain. The moisture in the upper 6 feet of soil was reduced below the wilting coefficient about June 20. The trees were showing evident signs of distress at this time and some of the leaves were dropping. As the season advanced, defoliation continued until about one-half of the leaves had dropped from the trees by September 5.

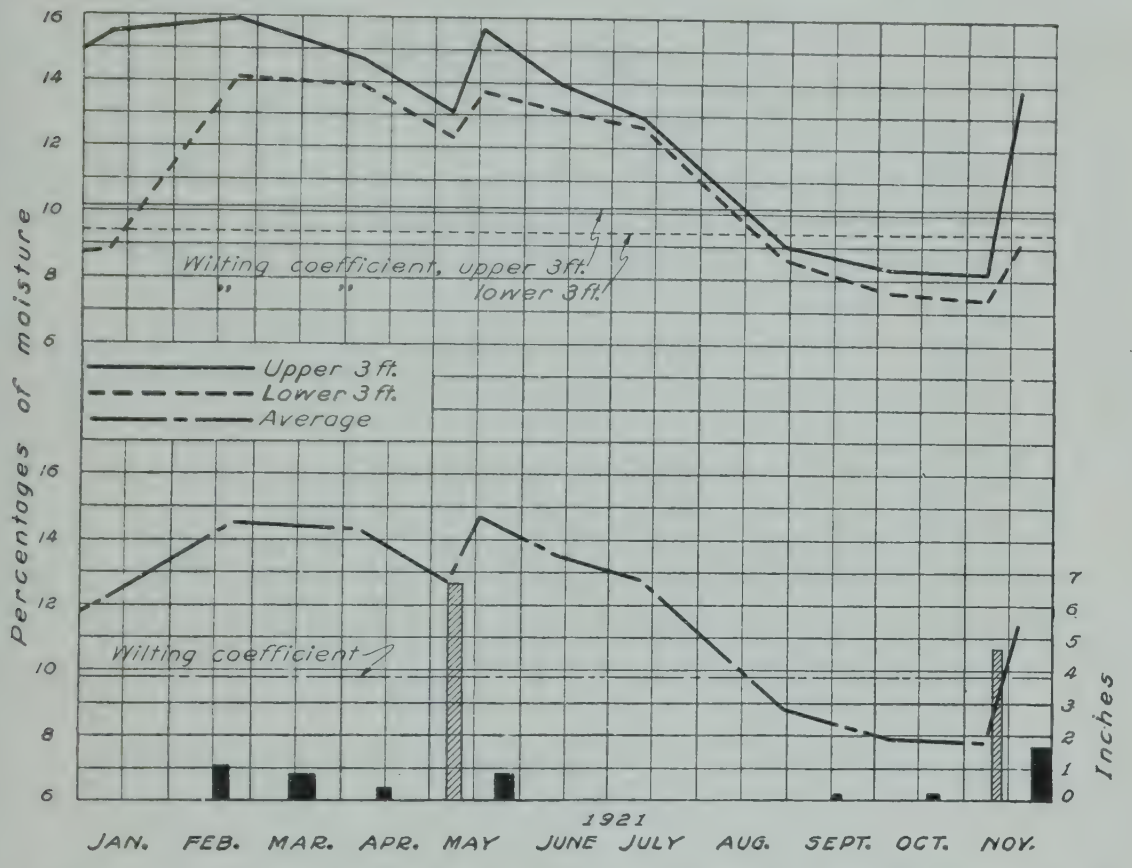


Fig. 5. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 1 during 1921. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

The heavier application of water later in the season of 1921 resulted in prolonging the period in which the soil-moisture supply in orchard No. 1 was above the wilting coefficient. However, the trees were badly wilted by August 30 and were practically defoliated by October 4.

The soil-moisture history of orchard No. 2, illustrated in figures 6 and 7, shows that the moisture content of the upper 6 feet of soil in this orchard was reduced below the wilting coefficient in each of the four years between July 1 and July 15. The orchard showed the lack of water during each growing season. Permanent wilting was evidenced at these times by wilting and shedding of the leaves.

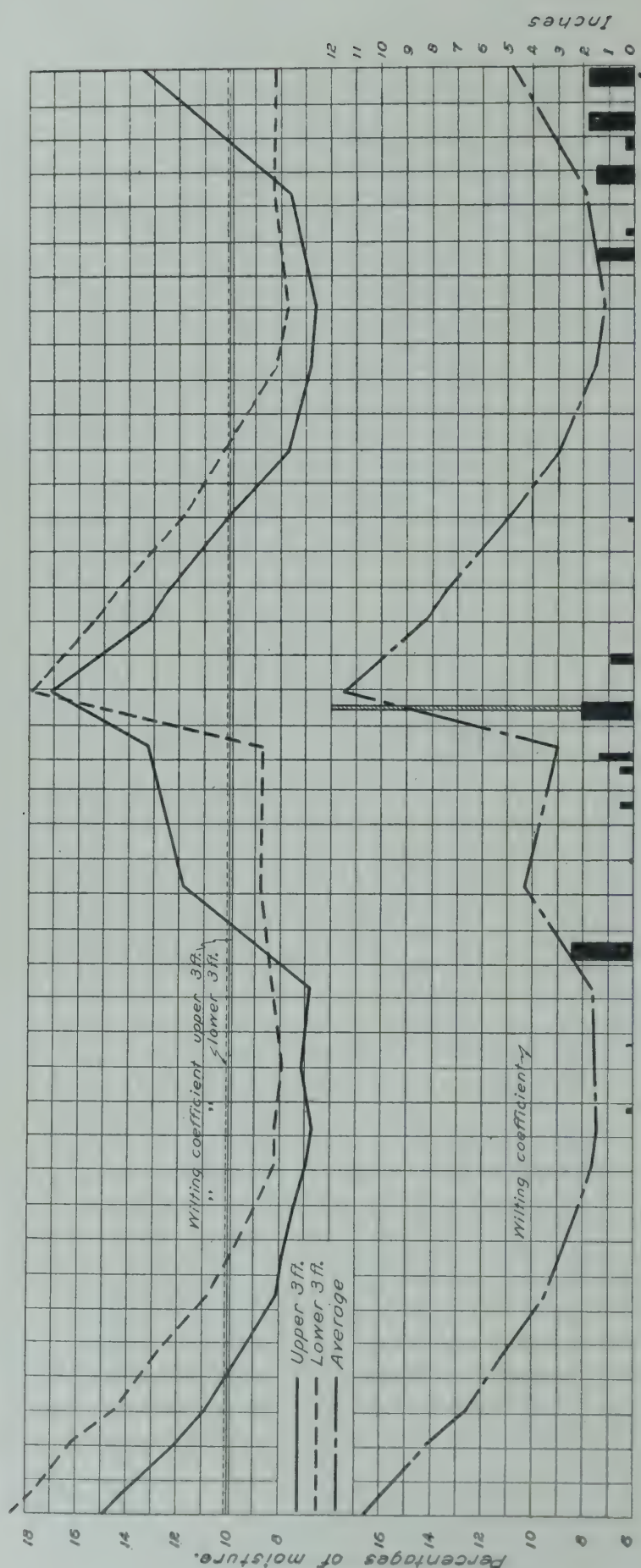


Fig. 6. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 2 during 1919 and 1920. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

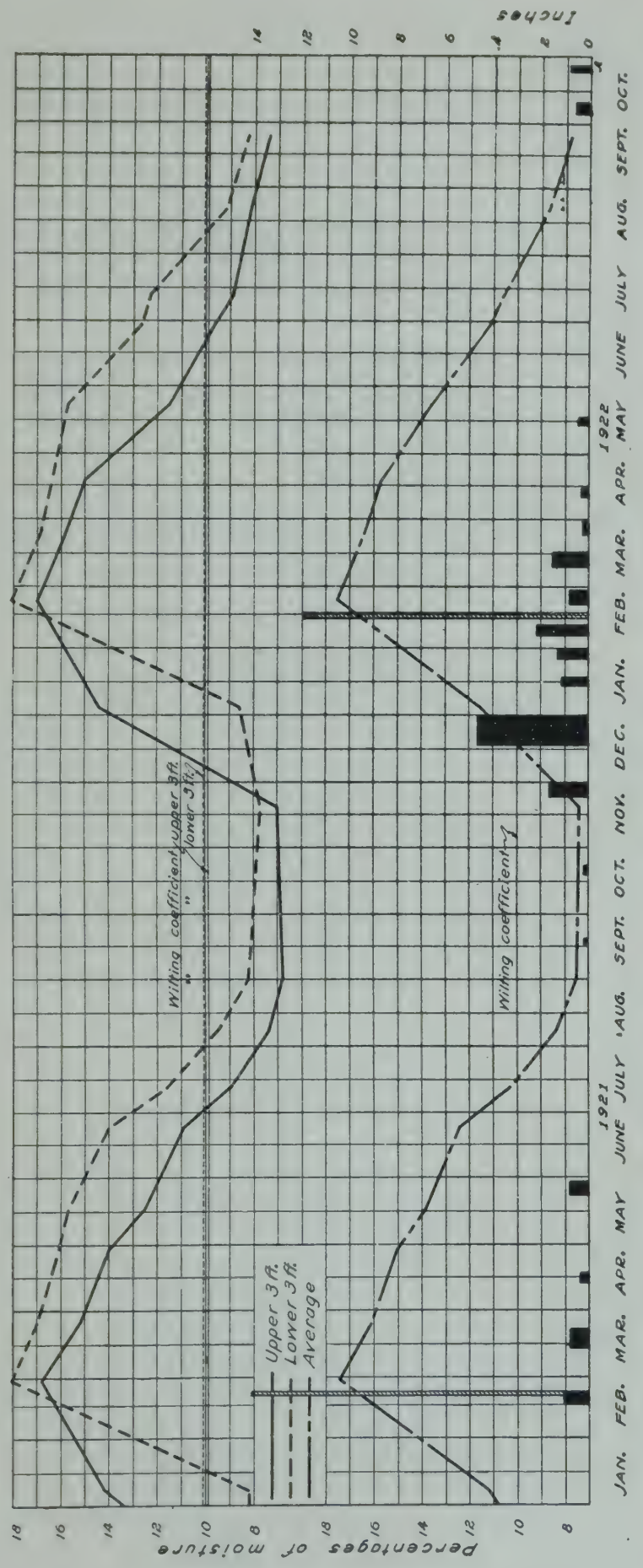

Percentages of moisture
Inches
Upper 3 ft.
Lower 3 ft.
Average
Wilting coefficient upper 3 ft.
" " lower 3 ft.
Wilting coefficient
1921
1922
JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT.

Fig. 7. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 2 during 1921 and 1922. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

The usual condition of the trees in this orchard after the removal of the crop is illustrated in figure 1. It will be noted that the trees were practically defoliated by October 4, 1920. This orchard, except during 1922, bore exceptionally good crops of fruit. The records of yields of fresh fruit from this orchard for the several years preceding 1919 are as follows: 1916, 8,467 pounds to the acre; 1917, 10,956 pounds to the acre; and 1918, 7,969 pounds to the acre. It is, indeed, remarkable that an orchard which unmistakably suffers from lack of water for such long periods of time during each year consistently bears good crops of fruit.

The graphs of the moisture condition in orchard No. 2, when compared with those illustrating conditions in the other orchards which were not winter irrigated, suggest that irrigation in the dormant season does not materally postpone the time in the following growing season during which the moisture supply in the first 6 feet of soil is depleted below the wilting coefficient. A similar condition was found by Batchelor and Reed[6] with mature walnut trees.

On June 20, 1919, the trees in orchard No. 3 showed wilting of leaves resulting in some abscission. The leaves continued to drop as the season advanced, the trees being completely defoliated by October 10. As indicated in figure 8, the moisture supply of the soil to a depth of 6 feet was reduced by the middle of June, 1919, below the wilting coefficient. The unusually low drying ratio of fresh fruit to dried fruit of 1.69 for this year was doubtless due to the partial drying of the fruit on the trees. The condition of the trees during the years 1920, 1921, and 1922 was much better than in 1919. Wilting did not occur so early, and the trees were not defoliated until later in the season. However, the moisture supply in the first six feet of soil was below the wilting coefficient for about two months during the latter part of the growing season each year. (Figures 8 and 9.) The trees were wilted during these periods and were revived on the application of water in the fall. This was especially noticeable in the years when the water was applied early in the fall, and before the majority of the leaves had dropped from the trees.

The yield from orchard No. 3, although much lower than any of the other orchards, was somewhat better than had been obtained for the several preceding seasons. The increase in yields after 1919, as shown in table 3, is a response to better care and to an available supply of soil moisture for a longer time each year.

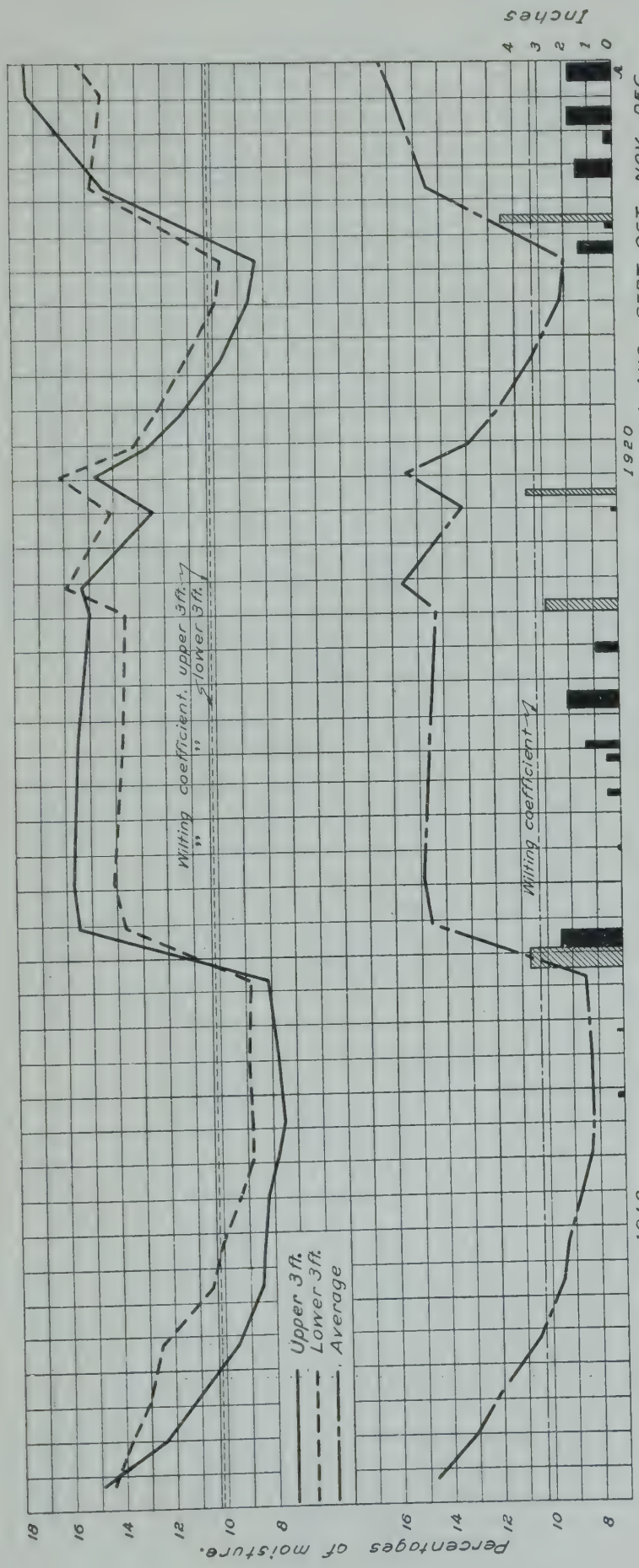


Fig. 8. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 3 during 1919 and 1920. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

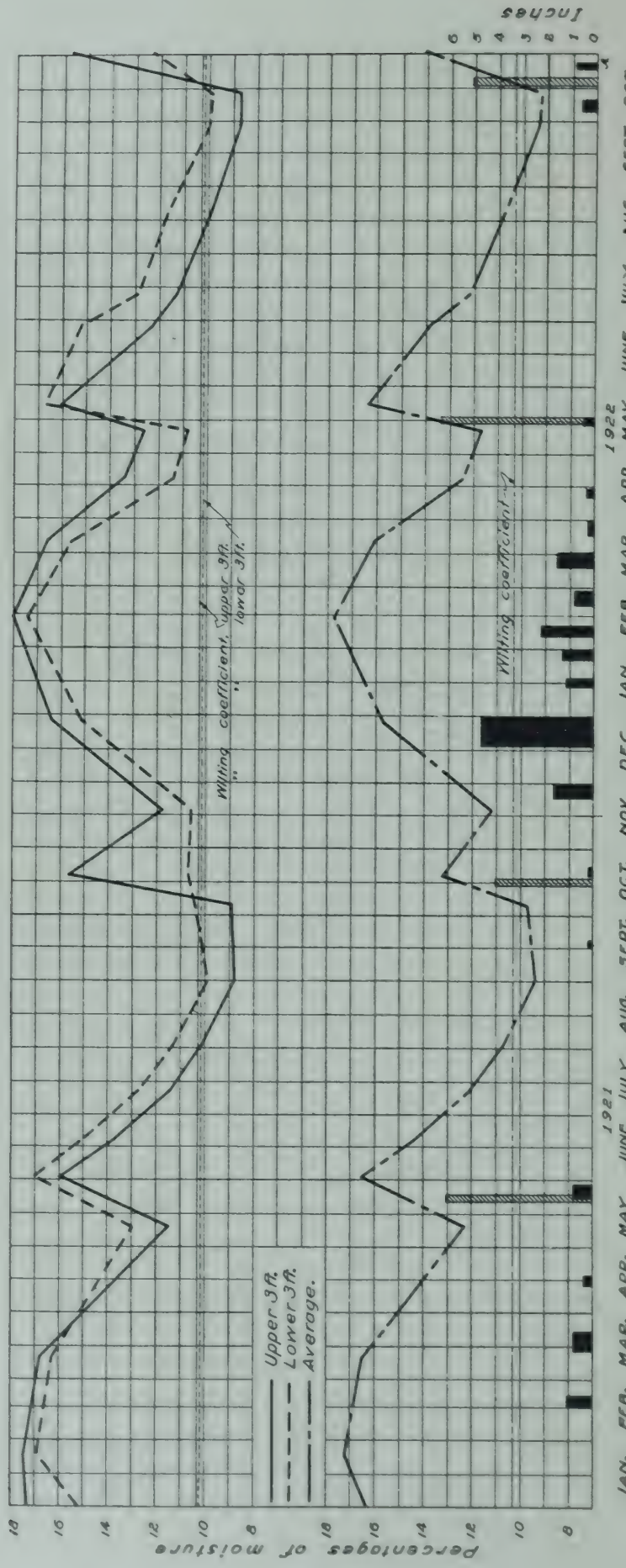

Inches
Percentages of moisture
Upper 3 ft.
Lower 3 ft.
Average.
Wilting coefficient. Upper 3 ft.
Lower 3 ft.
Wilting coefficient
JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT.
1921
1922

Fig. 9. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 3 during 1921 and 1922. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

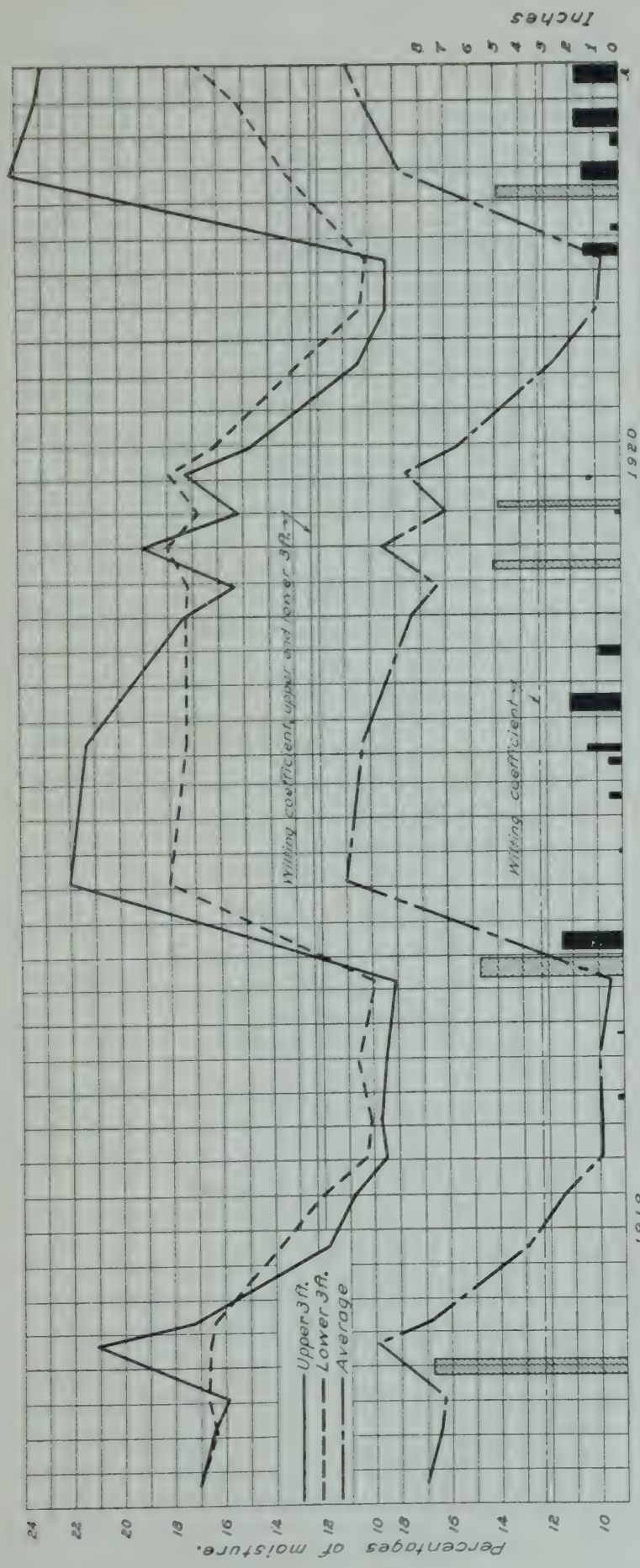

Inches
Percentages of moisture.
Upper 3 ft.
Lower 3 ft.
Average
Wilting coefficient, upper and lower 3 ft.
Wilting coefficient
1919
1920
APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC.

Fig. 10. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 4 during 1919 and 1920. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

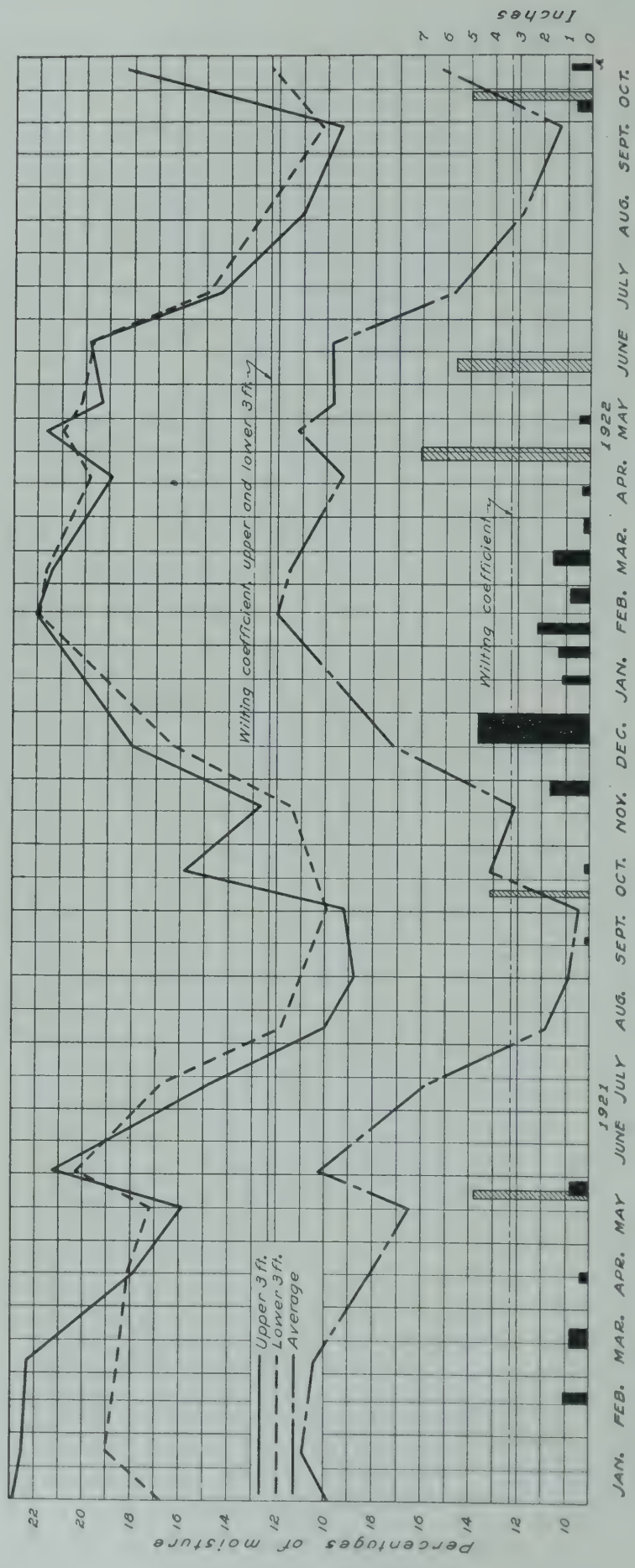

Inches
Percentages of moisture
Upper 3 ft.
Lower 3 ft.
Average
Wilting coefficient upper and lower 3 ft.
Wilting coefficient
1921
1922
JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT.

Fig. 11. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 4 during 1921 and 1922. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

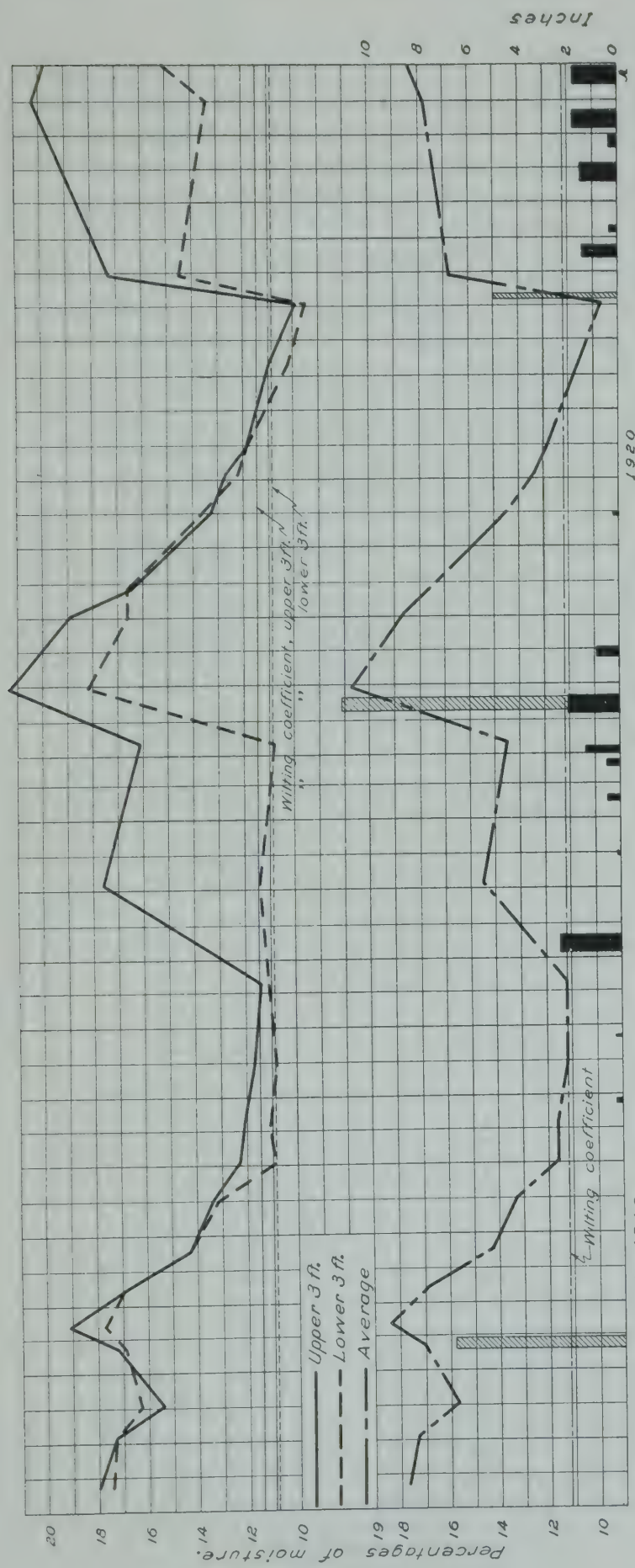

Inches
Percentages of moisture.
Upper 3 ft.
Lower 3 ft.
Average
Wilting coefficient, upper 3 ft.
" " lower 3 ft.
Wilting coefficient
1919
1920
APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC.

Fig. 12. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 5 during 1919 and 1920. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

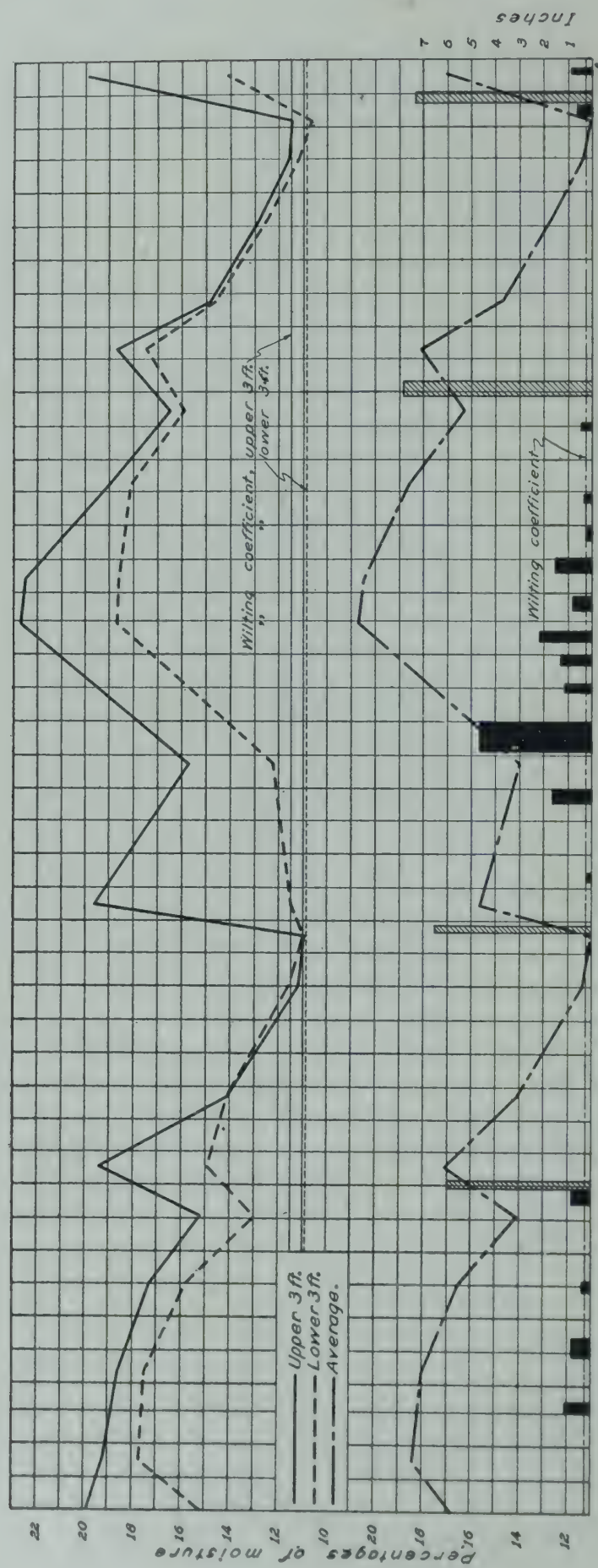

Inches
Upper 3 ft.
Lower 3 ft.
Average.
Wilting coefficient, upper 3 ft.
" " lower 3 ft.
Wilting coefficient
Percentages of moisture
1921
1922
JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT.

Fig. 13. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 5 during 1921 and 1922. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

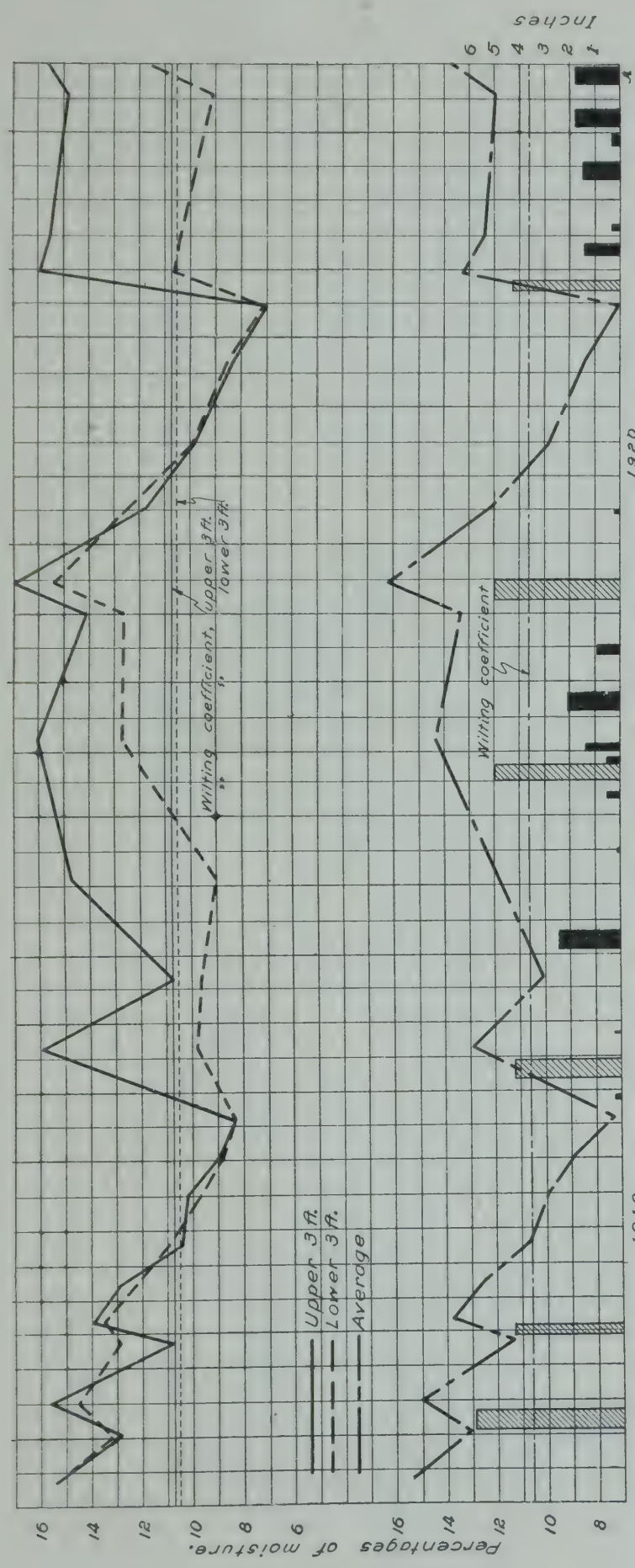

Inches
Percentages of moisture.
Upper 3 ft.
Lower 3 ft.
Average
Wilting coefficient, Upper 3 ft.
Lower 3 ft.
Wilting coefficient
1919
1920
APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC.

Fig. 14. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 6 during 1919 and 1920. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

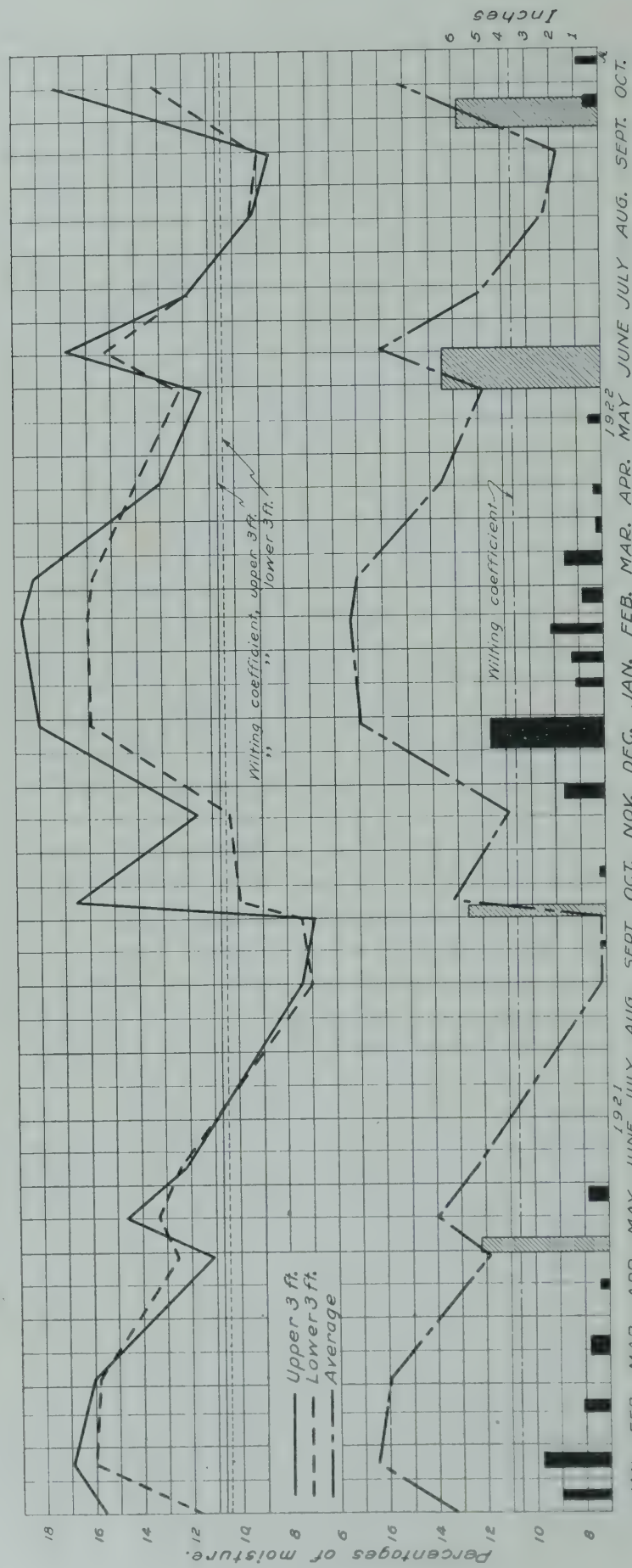

Percentages of moisture.
Inches
Upper 3 ft.
Lower 3 ft.
Average
Wilting coefficient, upper 3 ft.
" " lower 3 ft.
Wilting coefficient
1921
1922
JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT. NOV. DEC. JAN. FEB. MAR. APR. MAY JUNE JULY AUG. SEPT. OCT.

Fig. 15. Soil-moisture conditions in the Santa Clara Valley prune orchard No. 6 during 1921 and 1922. The rainfall is indicated by the heights of the solid black rectangles, and the amount of irrigation water is represented by the cross-hatched rectangles.

Orchards 4 and 6 had the moisture content of the first 6 feet of soil above the wilting coefficient for longer periods during the growing season than orchards 1, 2, and 3, as will be seen from figures 10, 11, 14, and 15. During the four years these orchards were under observation, the moisture content of the upper 6 feet of soil was below the wilting coefficient for periods of from 2 to 4 months during the growing season. These orchards wilted each year, and many of the leaves dropped before the crop was harvested. The condition of the trees in orchard No. 4 on November 1, 1920, is shown in figure 3. This condition was typical also of orchard No. 6. The trees apparently suffered from drought in spite of one or two rather heavy applications of water in the fore part of the season.

The soil-moisture history in orchard No. 5 is shown in figures 12 and 13.

Orchard No. 5 had better moisture conditions in the first 6 feet of soil throughout the four years than any of the other orchards. In one year only, 1920, did the moisture content fall below the theoretical wilting coefficient. In the latter part of the growing seasons of 1919 and 1922, the moisture content was reduced to an amount which was about equal to the wilting coefficient. It is interesting to note that some distress was indicated by the permanent wilting of some of the leaves. It is shown in table 4 that with the exception of the year 1920, this orchard had a higher yield to the tree than any of the other orchards. The striking difference in condition of the trees in orchard No. 4 and of those in orchard No. 5 could be noted easily since they were adjoining orchards. The border line of orchard No. 5 could clearly be distinguished from that of orchard No. 4 by the better appearance of the trees.

Orchard No. 5 had yielded good crops consistently before 1919. In 1916 the yield of fresh fruit to the acre was about 11,000 pounds; in 1917, 7,000 pounds; in 1918, 9,000 pounds. Orchard No. 4 had not been a good producer, the average yield of fresh fruit previous to 1919 having been about 4,400 pounds to the acre. Orchard No. 6 had not produced good crops up to 1919. The yield of fresh fruit to the acre in 1915 was 2,600 pounds, and in 1916, 6,435 pounds. In 1917 it was reduced to 2,918 pounds.

Extraction of Moisture by the Tree Roots from Depths of Soil Below Six Feet

Samples of soils for moisture determinations were taken to depths of 12 feet during the four years of observation. In each orchard the soil below 6 feet contained considerable gravel; the labor of taking samples, especially when the soil was dry, was so great that only a few sets of samples were taken each season. These determinations, though few in number, indicate that in every year during which soil moisture was available, some moisture was taken from the soil by the trees to the full depth of 12 feet. The extraction of moisture from the depth of 6 to 9 feet was at a slower rate than that from the first 6 feet. The extraction from the 9 to 12-foot depth was at a slower rate than from the 6 to 9-foot depth. The amounts of water taken from the soil by the trees from the four 3-foot layers of soil are given in table 5. These records show the extraction of moisture after the last irrigation was applied in the summer, or after the last rains, and before the first application of water in the fall, or at the end of the growing season. The values, expressed in acre-inches, are the amounts of water in 3 feet of soil, equivalent to the designated depth of water in inches. The amount of water in the soil in the fore part of the summer is given as the amount above the calculated hygroscopic coefficient. The values will convey some idea of the supply of water available for growth. However, it is to be noted from table 5 that in one instance only all of the water above the hygroscopic coefficient was used by the trees. This is the 0 to 3-foot depth of soil in orchard No. 6, during the summer of 1921. There was an amount of water equivalent to 3.68 acre-inches to the acre in this depth of soil on May 16, 1921, and 3.88 acre-inches to the acre was removed by the trees. The soil-moisture content in this orchard also was reduced to the hygroscopic coefficient in the fall of 1920. The moisture content of the upper 3 feet of soil in orchard No. 2 was usually reduced almost to the calculated hygroscopic coefficient in the fall of each year.

The smaller amounts of water taken from the 6 to 9-foot depth, and the still smaller amounts taken from the 9 to 12-foot depths of soil, is clearly brought out in table 5. These records, together with the graphs of moisture conditions, indicate that if the losses of moisture from below the surface layers of soils are attributed solely to that caused by transpiration through the trees, an assumption

TABLE 5

AMOUNTS OF MOISTURE TAKEN BY TREES FROM DIFFERENT DEPTHS OF SOIL BETWEEN IRRIGATIONS IN SUMMER AND FALL*

Orchard	Date of sampling	Acre-inches per acre available above hygroscopic coefficient				Depth of soil wet from previous irrigation	Samples taken before irrigation. Date	Acre-inches to the acre taken from different depths of soil				Percentage taken of total amount available			
		0-3 foot depth	3-6 foot depth	6-9 foot depth	9-12 foot depth			0-3 foot depth	3-6 foot depth	6-9 foot depth	9-12 foot depth	0-3 foot depth	3-6 foot depth	6-9 foot depth	9-12 foot depth
1	May 16, 1921	4.39	3.68	2.84	2.47	10.5 feet	Oct. 4, 1921	3.86	3.07	2.32	1.81	87.8	83.5	81.8	73.3†
2	Apr. 28, 1921	3.63	4.64	4.29	3.03	Below 12 feet	Nov. 18, 1921	3.53	4.24	3.35	1.26	97.2	91.4	78.0	41.6
2	Apr. 17, 1922	4.13	4.79	4.84	4.18	Below 12 feet	Sept. 28, 1922	3.83	4.08	3.73	1.59	92.7	85.3	78.7	38.9
3	June 1, 1921	4.54	5.04	3.56	2.52	9.5 feet	Oct. 4, 1921	3.58	3.33	2.55	1.71	78.8	66.1	71.7	67.8†
3	June 28, 1922	2.82	4.14	2.72	1.64	10.5 feet	Sept. 28, 1922	1.97	2.62	1.49	1.17	69.9	63.3	56.8	71.3†
4	June 1, 1921	6.50	6.04	4.78	2.67	Below 12 feet	Oct. 1, 1921	6.04	5.74	3.27	0.60	93.0	95.0	68.4	22.5
4	June 28, 1922	5.80	5.74	4.69	4.79	Below 12 feet	Sept. 27, 1922	5.24	4.29	1.94	0.98	90.2	74.7	41.3	20.5
5	June 19, 1922	5.50	5.09	3.68	2.92	Below 12 feet	Sept. 15, 1922	3.58	3.07	1.92	1.16	65.1	60.4	52.2	39.7
6	May 16, 1921	3.68	3.13	1.87	1.18	6.5 feet	Oct. 1, 1921	3.88	2.97	1.56	1.06	105.4	94.9	83.5	89.9†
6	June 17, 1922	4.79	4.08	2.97	1.56	9.0 feet	Sept. 14, 1922	4.18	3.13	2.17	1.21	87.4	76.6	73.1	77.5†

* The water table under each of these orchards was over 100 feet from surface.

† It should be noted that the soil from 9 to 12 feet was not fully wet from previous irrigation.

which will be shown to be substantially true in the latter part of this report, most of the roots are distributed rather uniformly throughout the upper 6 feet of soil, there are a few roots in the 6 to 9-foot depth, and still fewer in the 9 to 12-foot depth.

In orchard No. 4, very little moisture was taken from the 9 to 12-foot depth even though there were large amounts available. The soil to this depth was noticeably moist at all times during the last three years of the period of observation. In spite of this, the trees in this orchard showed permanent wilting and defoliation of leaves during the growing season. This suggests that the distribution of Myrobalan roots, on which these trees were grown, was largely limited to the upper 9 feet of soil.

Discussion of Results

The graphs of the soil-moisture conditions in the Santa Clara Valley prune orchards show the fluctuations in soil moisture prevalent in mature commercial orchard. These suggest that the maintenance of even an approximately uniform soil-moisture content is impossible. Mature prune orchards are different from other deciduous orchards in that there is a long period when surface applications of water can not be made. Usually the trees are propped for about two months during the latter part of the summer, and the fruit is on the ground for a long time.

Orchard No. 5 had the best moisture conditions of any of those under observation. An irrigation was given in June of each year, except in 1920, and the orchard was irrigated as early as possible in the fall. The soil moisture was reduced appreciably below the wilting coefficient only in 1920. Orchards No. 4 and No. 6 showed greater reductions of the soil-moisture supply than No. 5. Orchards No. 1 and No. 2 were wilted for longer periods than the other orchards, except No. 3, during 1919 and 1920. Orchard No. 3 showed much less wilting during the season of 1921 and 1922. It ranks with orchard No. 4 in this respect.

There seems to be no relation between the yield and the soil-moisture content in the different orchards. Also the size of dried prunes bore no relation to soil-moisture content. The number of pounds of fresh fruit required to make one pound of dried fruit, given in table 4, can not be used to judge strictly the amount of water in the fresh fruit from the different orchards. The time the fruit was allowed to remain on the ground and the condition of

dryness of the fruit probably were not the same for the different orchards. As previously pointed out, the low drying ratio for the fruit from orchard No. 3 in 1919 was due to the fact that much of the fruit dried on the trees. However, the drying ratios recorded represent what may be expected from commercial orchards and indicate no consistent relation between soil moisture and water content of the fruit. This is further illustrated in the percentages of water found in the flesh of the fruit samples collected from the different orchards and reported in table 4. As previously discussed, it is also thought that the differences in amounts of sugar found in these samples can not be attributed to the differences in irrigation.

No differences could be recognized in the condition of trees growing on soil with a high moisture content in the upper 6 feet and those on soils with low moisture content in the same depth until the soil-moisture supply had been reduced to the wilting coefficient. Wilting, without recovery until water was again added to the soil, always resulted when the moisture content of the upper 6 feet of soil had been reduced to a condition closely approximating the calculated wilting coefficient. This will be discussed further in the following sections.

Since the trees did not seem to be affected until the soil-moisture content had been reduced to the wilting coefficient, the necessity for irrigation early in the summer, when there was considerable residual soil moisture either from rains or previous irrigations, is not apparent. The irrigation practice in all of these prune orchards could have been bettered if the spring irrigations were delayed and the water applied later in the season when the moisture content of the upper 6 feet of soil was further reduced. Many of the early irrigations resulted in penetration of water below the 6-foot depth. The same amount of water applied later would delay wilting and shorten the time the trees would remain in this condition. The soil moisture more nearly approached this condition in orchard No. 5 than in any of the other orchards.

The graphs depicting the soil-moisture conditions (figures 4–15) do not indicate a more rapid use of water by the trees on soil with high moisture content than by those on soil with low moisture content. This is evidenced by a comparison of the slopes of the portion of the graphs following irrigation with the portions near the wilting coefficient.

Some of the graphs do show a steeper slope immediately following the application of water. For example, the graphs in figure 10 for

the 0 to 3-foot depth and the 0 to 6-foot depth are steeper following the irrigation of May 27 to June 3, 1919. However, this as well as other cases which can be noted in the graphs may be due to the rapid loss by evaporation from the surface layer of soil immediately following irrigation and to downward movement which carried some of the water below the upper 6 feet of soil. In many cases soil samples were taken immediately after irrigation and before the water was distributed throughout the soil. The samples included the surface layer from which rapid loss from evaporation followed irrigation. The loss of water from below the surface of the soil is assumed to be almost entirely due to transpiration, and the slope of the graphs to indicate the rate of extraction by the roots. These considerations will be more fully dealt with in later sections of this report. The rate of extraction of soil moisture seemed to be as rapid from the 0 to 3-foot depth of soil as it was from the 3 to 6-foot depth.

The condition of the trees seemed to be governed by the moisture content of the upper 6 feet of soil. When the moisture in this depth of soil had been reduced to the wilting coefficient, wilting always resulted and the trees did not recover until water was added to the soil, even though the soil below the sixth foot had a much higher moisture content.

Further information concerning the use of water by mature deciduous fruit trees is given in the following section.

SECTION II

IRRIGATION STUDIES OF PEACHES

Studies similar to those made in the Santa Clara prune orchards were made on the effect of irrigation on peaches. An orchard consisting of one block of Muir peaches, on peach root, located at the Branch of the College of Agriculture of the University of California, at Davis, was selected.

This locality is a semi-arid region. There are no fogs during the growing season, and between the first of May and the latter part of September there are few clouds. The area is typical of the hot, dry interior-valley climate of California, except that the night temperatures are lower than are usual farther north in the Sacramento Valley and in the southern part of the San Joaquin Valley. The normal rainfall at Davis, calculated up to 1923, is 17.41 inches. Practically no rain falls during the summer months. During the six years these studies were conducted, the rainfall was less than normal except during the season of 1918–1919. The rainfall in inches given in the following tabulation, is for each season from September 1 of one year to September 1 of the following year.

1916–1917	14.09	1919–1920	8.98
1917–1918	9.66	1920–1921	17.13
1918–1919	19.40	1921–1922	16.63

The soil in this area is classed as Yolo loam. In some places the surface soil verges into a silty loam or a fine sandy loam. The subsoil is somewhat lighter in texture than the surface soil, and there are occasional pockets of sand and gravel.

The distance to the underground water surface under the orchard during the years the observations were made was about 18 feet. The normal rainfall at Davis is usually sufficient to wet the soil, if it is dry in the fall, to a depth of about 8 feet. Results of soil sampling indicate that this moisture is exhausted by mature peach trees at the end of the growing season of each year. It is possible that the roots of the trees, in the area under observation, were suf-

ficiently deep to extract moisture from the moist soil immediately above the water-table. However, in the principal peach growing sections of California, the water-table is generally higher than that at Davis, and it was thought that the results obtained in a study of the effect of the variation of the moisture supply in the upper 6 feet of soil, would be applicable to other peach growing areas in the State.

On February 16, 1912, the trees, which were excellent two-year-old nursery stock, selected for uniform size, were planted in the orchard. The irrigation and cultural treatments were the same throughout the orchard until the beginning of the growing season of 1917, when differential irrigation treatments began.

The tree rows were numbered 1, 2, 3, ... beginning on the south side, and so on to 16 on the north end. The trees in each row were numbered 1 to 6, beginning with number 1, on the east side, and ending with number 6, on the west side. The trees were spaced 20 feet in the rows and the rows were 20 feet apart.

The schedule of irrigation treatment which was followed during 1917 and 1918 was as follows: All even numbered rows (rows 2, 4, 6, 8, 10, 12, 14, and 16) received no irrigation when there was normal rainfall. The normal rainfall at Davis, up to the year 1917, from September 1 to January 1, was 6.01 inches; to February 1, 9.71 inches; to March 1, 12.53 inches; and to April 1, 15.14 inches. Whenever the rainfall was below normal on these dates, sufficient water was applied to make up the deficiency.

Rows 1 and 9 received no irrigation.

Rows 3 and 11 were irrigated in the fore part of growing season (April or May, depending upon rainfall). They were also irrigated shortly before harvesting and about 30 days after harvesting.

Rows 5 and 13 were irrigated sufficiently to insure a high soil-moisture content at time of blossoming (February or March, if rainfall were not sufficient), at the time of rapid length growth (April or May), and shortly before harvesting.

Rows 7 and 15 were irrigated to insure ample moisture in the soil at the time of most rapid fruit growth (May or June), and about 30 days after harvesting.

The cultural treatment was the same throughout the orchard during the entire period of observation. The orchard was plowed early in the spring of each season, in order to kill all volunteer vegetation. No cover crops were planted. The plowing was followed by disking and harrowing, and additional cultivations were given

only to keep down weed growth, and to prepare the soil before and after irrigation. Pruning of the different rows was kept as uniform as possible. The recorded weights of pruning from the different rows showed no appreciable differences in the amounts of wood removed from the different trees. Thinning was done as uniformly as possible. No fertilizer was added to the soil, and it is believed that the only material difference between the rows was the amount of moisture available in the soil.

The amount of water to be applied at the times specified in the above schedule was determined by sampling the soil to a depth of 6 feet. Whenever the moisture content of the soil in the rows which were to be irrigated according to the schedule previously given was found to be less than the maximum field capacity, the requisite amount of water was applied. The maximum field capacity of the soil was determined by applying an amount of water equivalent to 12 inches in depth, to 5 plots of about 100 square feet each, and taking 13 samples in each plot to a depth of 9 feet. The samples were not taken until after gravitational movement of the water downward had ceased. The plots were located so that variations in soil would be represented. The average water holding capacity of the upper 6 feet of soil was determined by this method to be 20.18 per cent.*

Sampling the soil during 1917 and 1918 was done with a post-hole type of soil auger. A hole was bored and a sample taken from each foot of soil. The subdivision of the sample was made in the field. As previously pointed out, this method of sampling is sometimes faulty, and inconsistencies in the results were observed frequently. It was found that the amount of water applied could not always be accounted for in the soil by this means of sampling after irrigation.

During the seasons of 1917 and 1918, about 6,000 samples were taken in the orchard. The results, in 1917, of sampling the soil to determine the moisture content, and the record of irrigations are given in table 6. For assistance in the interpretation of these data, the average moisture equivalents of the different depths of soil are given at the bottom of the table. Samples from the same rows were centrifuged at different times. However, the samples used for these moisture equivalent determinations were not exactly 30 grams. The values for the wilting coefficients and hygroscopic coefficients, calculated from those for the moisture equivalents, are also listed.

* This method of determining the amount of water the soil is capable of holding is subject to some objection since Israelsen and West[34] seem to show that the amount of water held by the soil is dependent, to some extent, on the amount of water applied.

TABLE 6

SUMMARY OF IRRIGATION TREATMENTS AND SOIL-MOISTURE CONTENTS IN MUIR PEACH ORCHARD, DAVIS, 1917

Row	Dates of irrigation	Depth of water applied in acre-inches to the acre	Dates of sampling	Percentages of moisture in the soil						Average percentage of moisture in 6 feet	Equivalent depth of water in inches
				1st foot	2nd foot	3rd foot	4th foot	5th foot	6th foot		
1, 9	.....	.....	Mar. 15	19.0	19.0	16.4	17.0	17.4	15.8	17.4	16.3
			Aug. 7	10.1	11.0	10.4	10.4	9.2	10.3	10.2	9.6
			Oct. 1	8.5	10.8	10.2	9.8	9.5	10.1	9.8	9.2
2, 4, 6	May 4–6.....	2.5	Mar. 15	19.4	18.3	16.4	17.0	17.2	15.1	17.1	16.0
8, 10, 12			May 2	16.9	15.9	15.7	17.6	19.7	17.1	17.2	16.0
14, 16			May 8	20.1	19.2	17.8	18.2	18.4	15.9	18.3	17.1
			Aug. 7	10.9	11.9	11.3	11.2	12.6	10.8	11.3	10.5
			Oct. 1	9.4	10.8	10.1	9.7	9.6	9.5	9.7	9.0
3, 11	May 18–21..	5.33	Mar. 15	18.1	17.7	16.8	16.2	16.5	14.5	16.6	15.5
			May 15	12.7	15.7	15.4	16.2	14.8	14.3	14.9	13.9
	Aug. 13–16	8.05	May 22	19.8	19.6	18.0	18.1	18.2	16.6	18.4	17.2
			Aug. 7	11.8	12.1	11.4	11.4	12.0	10.7	11.6	10.8
	Oct. 8–13....	7.76	Aug. 21	19.5	17.6	16.2	17.1	16.6	14.6	16.9	15.8
			Oct. 1	13.7	12.0	11.9	12.0	11.9	9.3	11.9	11.1
			Oct. 18	21.1	18.7	17.1	16.7	16.0	14.0	17.1	16.0
5, 13	May 19–21..	5.28	Mar. 15	18.9	18.0	16.6	17.3	19.0	16.1	17.7	16.6
			May 15	10.8	14.8	14.6	10.8	11.6	16.1	14.5	13.6
			May 22	20.8	19.4	18.0	15.7	16.8	14.6	17.6	16.4
	Aug. 13–16	8.41	Aug. 7	11.4	11.8	11.0	10.9	11.1	10.9	11.2	10.5
			Aug. 21	20.4	18.9	16.6	16.0	14.4	14.0	16.7	15.6
			Oct. 1	14.3	13.5	12.8	12.6	12.1	11.3	13.8	11.9
7, 15	July 2–3.....	6.17	Mar. 15	19.9	19.1	17.2	17.1	18.1	16.6	18.2	17.0
			July 2	13.0	13.1	12.5	13.8	15.1	14.0	13.6	12.7
			July 7	20.5	18.4	15.4	16.1	15.2	14.8	16.7	15.6
	Oct. 8–13....	9.00	Aug. 10	15.0	14.1	12.9	12.9	11.8	12.7	13.3	12.4
			Oct. 1	11.2	10.9	10.1	10.1	9.8	11.1	10.6	9.9
			Oct. 18	22.5	18.8	17.5	18.0	14.0	14.0	17.5	16.4
Moisture equivalent.....				23.5 ±0.12	21.8 ±0.18	19.3 ±0.27	18.5 ±0.40	18.5 ±0.50	17.9 ±0.67		
Wilting coefficient.....				12.8 ±0.06	11.8 ±0.10	10.5 ±0.14	10.1 ±0.22	10.0 ±0.27	9.8 ±0.36		
Hygroscopic coefficient.....				8.7 ±0.04	8.1 ±0.07	7.1 ±0.10	6.8 ±0.15	6.8 ±0.18	6.6 ±0.25		

TABLE 7

SUMMARY OF YIELDS FROM MUIR PEACH ORCHARD, DAVIS, 1917

Row	Number of trees	Dates irrigated	Total fresh fruit, including culls	Culls	Number mature peaches	Weight fresh fruit	Average weight to a fruit, fresh	Weight dried fruit	Number of pounds of fresh fruit to make one pound of dried fruit
			Pounds	*Pounds*		*Pounds*	*Pounds*	*Pounds*	
1	6	No irrigation	115.4	71.0	125	44.4	0.36	5.4	8.29
9	6		370.7	197.2	510	173.5	0.34	23.2	7.47
	Average to a tree...		40.5	22.4	53 ±8.5	18.1 ±2.5	0.35 ±0.05	2.4 ±0.4	7.88
3	6	May 18–21	402.8	220.6	485	182.2	0.38	20.0	9.12
11	5	Aug. 13–16....	351.2	171.9	455	179.3	0.37	22.6	7.95
		Oct. 8–13.....							
	Average to a tree...		68.5	35.6	85.5 ±13.9	32.9 ±3.5	0.38 ±0.04	3.9 ±0.4	8.54
5	5	May 19–21....	528.4	262.0	814	266.4	0.33	37.9	7.04
13	6	Aug. 13–16....	319.7	162.9	467	156.8	0.34	21.3	7.36
	Average to a tree...		77.1	38.6	116.5 ±21.4	38.5 ±6.8	0.33 ±0.06	5.4 ±1.0	7.20
7	6	July 2–3.....	512.5	265.9	660	246.6	0.38	31.7	7.78
15	6	Oct. 8–13.....	306.9	150.2	411	156.7	0.38	20.5	7.65
	Average to a tree...		68.3	34.7	89.3 ±9.3	33.6 ±2.9	0.38 ±0.03	4.4 ±0.4	7.72
2	6		409.3	185.7	595	223.6	0.38	28.2	8.03
4	5		380.4	194.4	504	186.0	0.37	23.3	7.97
6	6		410.8	211.5	606	199.3	0.33	29.1	6.85
8	5	May 4–6.....	237.4	113.3	329	124.1	0.38	16.0	7.89
10	4		186.4	81.9	211	104.5	0.49	15.2	6.85
12	6		390.0	199.0	538	191.0	0.35	26.3	7.25
14	6		286.6	149.9	391	136.7	0.35	18.2	7.46
16	4		92.1	34.6	139	57.5	0.41	6.4	9.03
	Average to a tree...		55.7	27.2	78.7 ±4.6	28.5 ±1.5	0.36 ±0.02	3.8 ±0.3	7.67

All irrigation water during the season of 1917 was applied by furrow irrigation, six zigzag furrows being used for each tree row. The water applied was measured by means of hook-gage readings on a "V" notch weir.

The first blossoms appeared on the trees on March 10, and March 18 was estimated as the mid-blossoming period. The samples taken on March 17 showed the soil to be amply moist; therefore, the irrigations in rows 5 and 13, which were scheduled, were not given.

Fruit harvest on all rows started on August 27 and continued until September 10. There were no apparent differences in the dates of ripening of the fruit on the different rows. A summary of the yields for the season of 1917 is given in table 7. The yields from trees which had been replanted, or were obviously subnormal, are not included in the table.

Results Obtained During the Season of 1918

The method of sampling and the general procedure was the same in 1918 as in 1917. The method of irrigation was changed from the furrow to the basin method. Levees were constructed so that each tree was inclosed in a square basin which could be supplied with water from a delivery flume. Therefore, a definite amount of water could be supplied to each tree, with more even distribution in the soil.

The rainfall, up to February 1, was only 2.15 inches. This was 7.56 inches less than normal. The even numbered plots were therefore irrigated on February 4 and 5. The dates of the irrigation, amounts of water applied, and soil moisture records are given in table 8.

Complete records for the yields of the trees were not obtained for the season of 1918, but the yields of representative trees in the different rows were secured and compared. No significant difference could be determined in weight or size of fruit from the various irrigated rows. The yields from the trees in rows 1 and 9 were smaller than those from the other trees. The soil-moisture supply in these rows was below the wilting coefficient by June 1.

The frequent irrigations given the different rows and the interference due to the penetration of the roots of the even-numbered check rows into the adjacent irrigated rows were possible reasons for lack of difference both in the appearance of the trees and the yields obtained.

TABLE 8

Summary of Irrigation Treatments and Soil-Moisture Contents in Muir Peach Orchard, Davis, 1918

Row	Dates of irrigation	Depth of water applied in acre-inches per acre	Dates of sampling	Percentage of moisture in the soil						Average percentage of moisture in 6 feet	Equivalent depth of water in inches
				1st foot	2nd foot	3rd foot	4th foot	5th foot	6th foot		
1, 9	Sept. 11–12*	4.0*	Apr. 1	18.9	18.5	14.2	14.2	11.3	11.7	13.1	11.7
			June 1	18.7	11.3	10.7	10.0	9.1	9.6	9.9	9.2
			Sept. 21	18.5	13.0	10.1	8.6	7.8	8.2	11.4	10.7
3, 11	May 21	6.05	Apr. 1	18.5	17.9	15.8	16.5	15.2	14.0	16.3	15.3
	Aug. 12	10.30	May 20	12.3	13.0	13.2	14.8	14.6	14.6	13.7	12.8
	Sept. 11–12	4.0*	May 22	26.8	21.1	18.3	17.2	16.4	15.6	19.2	18.0
	Sept. 25	6.30	Aug. 7	9.3	10.0	9.1	9.2	8.3	8.4	9.1	8.5
			Aug. 14	28.2	21.6	13.9	12.4	11.9	10.4	16.4	15.4
			Sept. 21	19.1	16.7	12.6	12.1	11.1	13.4	13.4	12.6
			Sept. 26	25.7	21.4	19.9	17.1	16.0	10.6	18.4	17.4
2, 4, 6	Feb. 4–5	8.55†	Feb. 1	13.1	10.4	10.1	9.9	10.1	9.5	10.5	9.8
8,10, 12			Feb. 11	20.8	19.1	16.5	15.5	14.9	12.0	16.5	15.4
14, 16	Sept. 11–12	4.0*	Apr. 1	18.9	18.4	16.2	17.6	17.4	15.5	17.3	16.2
			June 1	9.9	12.7	13.0	14.6	14.1	14.1	13.1	12.2
			Sept. 21	20.5	14.8	10.4	9.8	9.3	8.2	12.2	11.4
5, 13	Apr. 4	5.95	Apr. 1	17.2	17.2	14.6	11.8	10.9	11.0	13.8	12.9
	May 21	4.5	Apr. 6	25.0	20.5	18.5	20.6	19.1	15.2	19.9	18.6
	Aug. 12	10.1	May 20	12.7	13.5	13.4	16.1	17.2	18.2	16.9	15.6
	Sept. 11–12	4.0*	Aug. 7	9.2	10.0	9.5	9.2	8.9	8.8	9.4	8.8
			Aug. 14	26.6	19.4	15.2	10.8	9.7	10.4	15.4	14.4
			Sept. 21	21.2	18.4	14.2	12.7	10.7	12.2	14.9	13.9
7, 15	July 2	9.78	Apr. 1	20.6	19.1	17.1	17.8	18.4	15.1	18.0	16.8
	Sept. 11–12	4.0*	June 1	8.4	11.3	11.7	13.4	13.9	12.4	11.8	11.0
	Sept. 25	7.38	June 28	10.6	10.3	10.0	10.2	8.6	8.7	9.8	9.1
			July 3	30.8	23.5	20.4	17.2	12.7	11.8	19.4	18.2
			Sept. 21	19.0	14.2	10.6	10.0	9.5	10.6	12.3	11.5
			Sept. 26	26.3	21.3	19.7	17.6	16.0	15.2	19.4	18.1

* Rainfall.
† Includes 0.99 inches of rain which fell during irrigation.
For moisture equivalents see table 6.

Results Obtained During the Season of 1919

Investigations of the root systems of the trees during the winter of 1918–1919 showed that the roots of the trees in adjacent rows overlapped, and there was a possibility that the trees in the check rows took moisture from the adjacent irrigated rows. For this reason the trees in the even-numbered rows were removed in February, 1919, leaving rows 1, 3, 5, 7, 9, 11, 13, and 15. This numbering was retained. The trees were then 20 feet apart in the rows and the rows were 40 feet apart. It was decided to give the two rows which constituted one treatment only one irrigation, instead of two or more irrigations which were given in the two previous seasons.

The differential irrigation treatments were as follows:

Rows 1 and 9 received no irrigation.

Rows 3 and 11 were irrigated near to the time when length growth was beginning to slacken. This was usually about the first week in June.

Rows 5 and 13 were irrigated shortly before harvesting. The time of application was about the first week in August.

Rows 7 and 15 were irrigated about 30 days after harvesting to observe the possible influence on dormancy and fruit bud formation. The irrigation was given about the middle of September.

These treatments were given during the remainder of the experiment which was carried on until the end of the season of 1922. It should be noted that these treatments are not essentially different from those given the same rows during 1917 and 1918. During 1917 and 1918, rows 1 and 9 were not irrigated; the soil moisture in rows 3 and 11 was maintained at a high percentage during the fore part of the season when rapid length growth was being made. The percentages of soil moisture in rows 5 and 13 were high during the time when the fruit was maturing; and rows 7 and 15 were supplied with ample moisture after the crop was removed.

The method used in 1917 and 1918 to determine the amount of water to apply, was followed in 1919 and subsequent seasons. At each irrigation, water was applied in amounts sufficient to raise the first 6 feet of soil to the maximum amount it was capable of holding against gravity. The trees were irrigated each time in basins, to insure the evenness of application of the water. Square basins 20 feet by 20 feet were constructed around each tree. Since the rows

TABLE 9

SUMMARY OF THE IRRIGATION TREATMENTS AND SOIL-MOISTURE CONTENTS IN THE MUIR PEACH ORCHARD, DAVIS, 1919

Row	Depth of soil sample	Dates irrigated	Depth of water applied in inches	Moisture content in percentage on dry weight basis																			
				Apr. 3	Apr. 18	May 22	June 5	June 7	June 9	June 11	July 11	Aug. 1	Aug. 6	Aug. 7	Aug. 11	Aug. 22	Aug. 26	Sept. 11	Sept. 15	Sept. 17	Sept. 20	Oct. 15	Nov. 25	Dec. 23
1, 9	0–3			15.4	14.8	11.6	10.5				9.2	8.6					8.7	8.6				8.6	8.7	13.0
	3–6			15.2	14.9	13.8	12.6				10.7	9.8					9.2	9.3				9 3	9.4	9.7
		Average		15.3	14.9	12.7	11.5				10.0	9.2					9.0	9.0				9.0	9.1	11.4
3, 11	0–3	June 6	8.06	16.0	14.9	12.2	11.0	23.2	20.6	18.2	12.1	10.7					9.6	9.4				9.5	9.4	14.3
	3–6			16.2	15.8	14.2	12.7	19.7	18.6	16.9	12.2	10.8					9.6	9.2				9.3	9.3	9.9
		Average		16.1	15.4	13.2	11.8	21.4	19.6	17.6	12.1	10.7					9.6	9.3				9.4	9.4	12.1
5, 13	0–3	Aug. 5	9.67	16.0	15.1	12.1	10.7				9.4	9.2	23.1	21.2	19.0	15.2	13.4	12.4				11.1	10.6	14.8
	3–6			15.8	15.0	13.7	12.4				9.8	10.3	18.2	18.6	16.6	14.9	13.2	12.2				11.0	10.4	10.5
		Average		15.9	15.0	12.6	11.5				9.6	9.8	20.6	19.9	17.8	15.0	13.3	12.3				11.1	10.3	12.7
7, 15	0–3	Sept. 11	10.21	16.2	15.2	11.8	11.1				9.6	9.2					8.8	8.7	22.8	20.1	18.2	17.0	16.8	18.4
	3–6			18.0	17.7	13.9	12.2				10.8	10.2					9.8	9.4	21.0	20.4	18.8	17.4	16.0	16.0
		Average		17.1	16.4	12.9	11.6				10.2	9.7					9.3	9.1	21.9	20.2	18.4	17.2	16.4	17.2

were 40 feet apart, this left a dry strip 20 feet wide between the rows. The interference of adjacent trees probably was greatly lessened by this method.

Beginning with the season of 1919, all soil samples were taken with a soil tube, and the results show much greater consistency in the moisture determinations. The samples were taken to a depth of 6 feet. The soil removed from the first 3 feet and that from the second 3 feet, were placed in separate soil cans. The results, in percentages, of moisture obtained were the averages of moisture contents of these two depths of soil. The entire core of soil removed from the tube was retained for the moisture determination. A summary of the irrigations applied and the moisture contents of the soil at different times throughout the season are given in table 9.

The moisture equivalents of the soil in the rows, grouped according to irrigation treatments, are given in table 10.

TABLE 10

SUMMARY OF MOISTURE EQUIVALENT DETERMINATIONS AND CALCULATED VALUES OF THE WILTING COEFFICIENTS AND HYGROSCOPIC COEFFICIENTS OF THE SOIL IN THE MUIR PEACH ORCHARD, DAVIS

Row		Depth of soil, in feet		
		0–3	3–6	0–6
1, 9	Moisture equivalent	18.82±0.38	16.24±0.40	17.53±0.27
	Wilting coefficient	10.23±0.21	8.82±0.22	9.33±0.16
	Hygroscopic coefficient	6.95±0.14	5.99±0.15	6.33±0.11
3, 11	Moisture equivalent	18.66±0.38	16.42±0.69	17.54±0.38
	Wilting coefficient	10.11±0.21	8.92±0.35	9.53±0.21
	Hygroscopic coefficient	6.89±0.14	6.06±0.25	6.47±0.14
5, 13	Moisture equivalent	19.60±0.23	16.00±0.50	17.80±0.26
	Wilting coefficient	10.65±0.12	8.70±0.27	9.68±0.14
	Hygroscopic coefficient	7.23±0.08	5.91±0.78	6.58±0.10
7, 15	Moisture equivalent	19.82±0.21	15.88±0.42	17.85±0.23
	Wilting coefficient	10.77±0.11	8.63±0.23	9.70±0.12
	Hygroscopic coefficient	7.31±0.08	5.86±0.15	6.59±0.08

The moisture contents of the soil in the rows throughout the growing season of 1919, are graphically illustrated in figure 16. The graphs in this figure are drawn in the same manner as those which illustrate the moisture conditions in the Santa Clara Valley orchards.

However, in these graphs, the scale is changed, the time for which the moisture contents are shown is shorter, and the losses of moisture from the soil following irrigation are more clearly defined. The lines representing the loss of moisture from the upper 3 feet of soil and those representing the loss from the lower 3 feet approach much closer

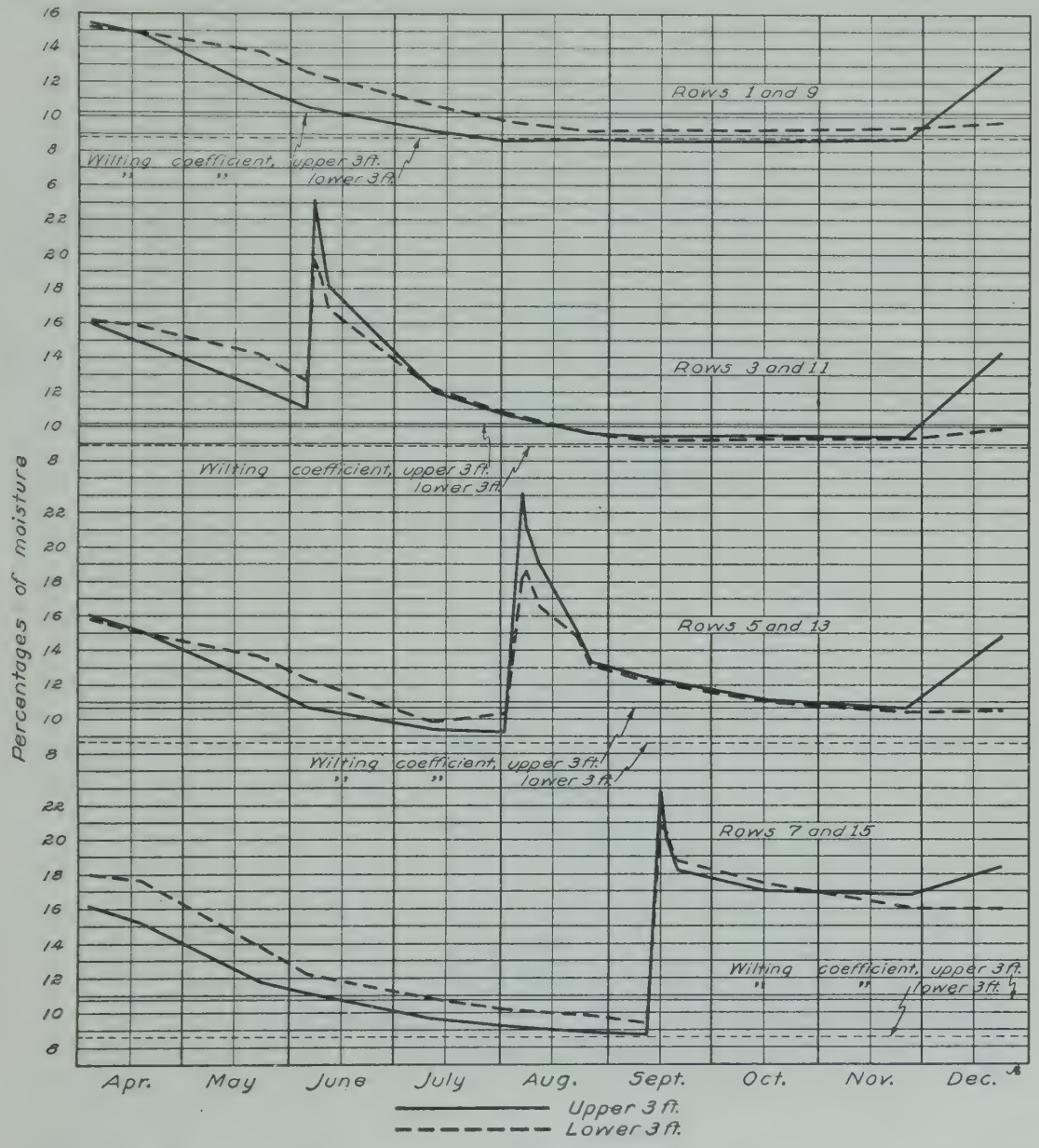


Fig. 16. Soil-moisture conditions in the Muir peach orchard at Davis during the growing season of 1919. The rows are grouped according to irrigation treatment.

to parallelism here than in the graphs of the soil moisture in the Santa Clara Valley orchards. At each irrigation in the peach orchard, sufficient water was applied to wet the soil to a full depth of 6 feet. This insured the same relative amounts of available water for growth in the two depths of soil.

The first picking was on August 13, 1919. Four pickings were made, the last one being made on August 27. There were no differences in the dates of ripening of the fruit in the different rows. A summary of the yields of fresh and dried fruit from the rows constituting the different treatments is given in table 11. These records show that there were no significant differences in the weight of fresh fruit, the weight of dried fruit, the number of fruits to the tree, the average weight of a peach; or the drying ratio.

After the peaches had been thinned, and after the chance of so-called "June drop" were lessened, measurements of circumferences of the peaches were started. Ten peaches, regularly spaced or located on each tree, were tagged and the maximum horizontal circumferences were measured with a small steel tape. The averages of these measurements are given in table 12. It is apparent from these records as well as those of the average weight of a peach given in table 11 that there were no significant differences in the sizes of peaches produced on the different trees.

TABLE 11

SUMMARY OF YIELDS FROM MUIR PEACH ORCHARD, DAVIS, 1919

Row	Number of trees	Dates irrigated	Total weight fresh fruit	Pounds culls	Number matured peaches	Weight fresh fruit	Average weight of a fruit fresh	Weight dried fruit	Number of fresh fruit to make one pound of dried fruit
1	6	No irrigation	1182.0	91.0	4249	1092.7	0.26	201.95	5.4
9	6		1308.5	97.5	5553	1211.0	0.22	235.30	5.2
	Average per tree.....		208.0	15.7	816 ±54.4	192.0 ±9.5	0.24 ±0.01	36.44	
3	6	June 5–6.....	1405.8	143.0	5105	1262.8	0.25	242.59	5.2
11	5		1093.5	143.5	3896	950.0	0.24	179.06	5.2
	Average per tree.....		227.0	26.1	820 ±38.1	201.0 ±6.8	0.25 ±0.01	38.33	
5	6	Aug. 4–5.....	1240.3	136.3	4448	1104.0	0.25	204.0	5.4
13	5		1143.0	166.5	4096	976.5	0.24	175.8	5.5
	Average per tree.....		216.6	27.5	775 ±56.6	187.0 ±12.8	0.24 ±0.02	34.5	
7	6	Sept. 11.....	1311.8	146.8	5391	1165.0	0.21	228.4	5.1
15	4		737.25	88.8	2505	648.5	0.25	131.5	4.9
	Average per tree.....		204.9	23.5	789 ±45.4	181.3 ±8.0	0.23 ±0.01	36.0	

Fig. 17. Wilting and partial defoliation of trees in the unirrigated row 9 caused by drought, August 18, 1920.

Fig. 18. Condition of trees in the unirrigated row 9 on August 2, 1919. Much less wilting is shown than in figure 17.

TABLE 12

AVERAGE SIZE OF PEACHES FROM MUIR PEACH ORCHARD, DAVIS, 1919. THE MAXIMUM HORIZONTAL CIRCUMFERENCE IN CENTIMETERS IS RECORDED

(Each value is the average of the measurements of 60* peaches for each row, or of 120 peaches for the average for each treatment.)

Row	June 21	July 11	July 30	Aug. 9	Aug. 15
1	12.4	14.0	16.5	18.7	19.3
9	11.7	13.0	15.4	17.6	18.2
Average	12.0 ±0.10	13.5 ±0.12	16.0 ±0.15	18.2 ±0.18	18.8 ±0.18
3	12.3	13.7	16.1	18.4	19.0
11	12.3	13.9	16.2	18.5	19.3
Average	12.3 ±0.07	13.8 ±0.09	16.2 ±0.13	18.4 ±0.13	19.2 ±0.19
5	12.2	13.6	16.1	18.5	19.2
13	11.9	13.3	15.7	18.5	19.3
Average	12.1 ±0.09	13.5 ±0.11	15.9 ±0.15	18.4 ±0.15	19.2 ±0.17
7	12.0	13.3	15.7	17.8	18.6
15	12.6	13.8	16.2	18.4	19.1
Average	12.3 ±0.09	13.6 ±0.10	16.0 ±0.10	18.1 ±0.17	18.8 ±0.16

* A few peaches dropped before the final measurements were made.

Fig. 19. Condition of trees in irrigated row 3 on August 2, 1919. The heavier foliage and absence of wilting is evident when this figure is compared with figures 17 and 18.

TABLE 13

SUMMARY OF IRRIGATION TREATMENTS AND SOIL-MOISTURE CONTENTS IN MUIR PEACH ORCHARDS, DAVIS, 1920

Row	Date of irrigation	Depth of water applied	Depth of soil in feet	Percentage of moisture								
				Apr. 5	Apr. 27, 29	June 5	June 16	July 10	Aug. 7	Aug. 10	Sept. 17	Sept. 24
1, 9	No irrigation		0–3	14.7	13.5	9.8		9.1	8.3		8.3	
			3–6	9.1	9.0	8.8		8.3	7.5		7.7	
			Average	11.9	11.3	9.3		8.7	7.9		8.0	
3, 11	June 9–10	9.96	0–3	14.9	15.2	9.9	17.1	12.5	10.8		8.4	
			3–6	9.3	11.3	9.0	15.4	13.3	9.4		8.8	
			Average	12.1	13.3	9.5	16.3	12.9	10.1		8.6	
3, 13	Aug. 7	11.04	0–3	15.0	13.4	10.2	10.1	9.2	8.5	22.2	10.9	
			3–6	9.2	9.4	9.1	9.2	9.2	8.2	19 0	13.6	
			Average	12.1	11.4	9.7	9.6	9.2	8.4	20.6	12.3	
7, 15	Sept. 20–21–22	10.68	0–3	17.7	15.9	10.4		9.8	8.7		8.4	21.6
			3–6	11.7	12.4	11.4		10.3	9.3		9.1	15.6
			Average	14.7	14.2	10.9		10.1	9.0		8.8	18.6

Results Obtained During the Seasons of 1920, 1921, and 1922

The procedure during the seasons of 1920, 1921, and 1922 was the same as that just described for the season of 1919. A partial summary of the soil moisture data of 1920 is given in table 13 for comparison with the soil-moisture condition in 1919, as shown in table 9. The rainfall during the winter of 1920 was only 8.35 inches, a deficiency of 6.79 inches. This was a particularly severe season for the unirrigated rows. The moisture content of the 3 to 6-foot depths of the soil in rows 1, 3, 5, 7, 9, and 11, was below the wilting coefficient on June 5. All of the trees in these rows were wilted at this time and some leaves had dropped. Row 9, the inside row which was not irrigated, clearly showed the effect of lack of water. The trees in row 1, the other unirrigated row, were not so badly wilted as those in row 9. The wilted and defoliated condition of the trees in row 9 on August 18, 1920, is illustrated by the photograph of tree 3 (fig. 17), which is typical of the trees in that row. While these trees showed some wilting in 1919 (fig. 18), they were in much better condition than in 1920. The condition of the trees in row 9 in 1919, can be further compared with the condition of those in row 3, by means of figures 18 and 19. The latter is a photograph taken of tree 3, in row 3, on August 2, 1919. The upper 3 feet of soil in row 3 had just been reduced to the wilting coefficient on August 1, but the lower 3 feet was above the wilting coefficient on this date.

Early in May a severe north wind blew for several days. This was an extremely hot and strong wind which caused much damage to the outside trees on the north and west sides of the orchard. For this reason the summary of the yields of only the inside trees in each row is given in table 14.

The first picking on all but row 9, was made on August 17, 1920, and the last picking on August 28. The fruit on row 9 did not mature until August 23. The last picking on this row was on September 3. Table 15 gives the average circumferences of peaches for the year 1920. These measurements were made in the same manner as those in 1919. The difference in size on the last date the measurements were made between the averages of rows 1 and 9, and 5 and 13, is 2.1 ± 0.55 centimeters. However, the size of the peaches in row 1 was not less than that of the peaches in row 3. The trees in row 9 were practically defoliated by the date of the last picking, September 3, and the peaches in this row were noticeably smaller than in the other rows.

TABLE 14

SUMMARY OF YIELDS OF INSIDE TREES OF MUIR PEACH ORCHARD, DAVIS, 1920

Row	Dates of irrigation	Number of trees in average	Weight of fresh fruit, pounds	Weight to a tree, pounds	Number of fresh fruit	Number to a tree	Average weight of a fruit, pounds	Weight of dried fruit	Weight of dried fruit to a tree, pounds	Pounds of fresh fruit for 1 pound of dried fruit	Weight of culls, pounds
9	No irrigation	4	238.5	59.5	1705	426	0.14	56.0	14.0	4.3	0
3, 11	June 9–10	7	797.0	114.0	3535	505	0.23	202.5	29.0	3.9	210.0
5, 13	Aug. 6	7	776.5	110.0	3572	510	0.22	176.5	25.0	4.4	86.0
7	Sept. 20–22	4	409.0	102.5	2205	551	0.19	95.0	24.0	4.3	35.0

TABLE 15

AVERAGE SIZE OF PEACHES FROM MUIR PEACH ORCHARD, DAVIS, 1920. THE MAXIMUM HORIZONTAL CIRCUMFERENCE IN CENTIMETERS IS RECORDED

(Each value is the average of the measurements of 60 peaches for each row, or of 120* peaches for the average for each treatment.)

Row	May 21	June 7	July 1	July 20	Aug. 6
1	10.3	11.5	13.1	16.0	18.7
9	10.9	11.2	12.8	14.6	16.8
Average	10.6 ±0.08	11.4 ±0.12	13.0 ±0.16	15.3 ±0.30	17.8 ±0.36
3	10.2	11.1	13.0	15.4	18.4
11	11.1	11.7	13.7	16.4	19.6
Average	10.6 ±0.10	11.4 ±0.12	13.4 ±0.13	15.9 ±0.18	19.0 ±0.25
5	10.6	13.3	15.2	18.2	20.6
13	11.3	12.1	14.0	16.4	19.2
Average	11.0 ±0.09	12.7 ±0.21	14.6 ±0.25	17.3 ±0.37	19.9 ±0.42
7	11.0	11.9	12.9	15.3	17.8
15	11.4	12.4	13.8	16.4	19.4
Average	11.2 ±0.05	12.2 ±0.10	13.4 ±0.19	15.8 ±0.19	18.6 ±0.32

* After August 6 so many peaches dropped that further measurements could not be made.

During the seasons of 1921 and 1922, the irrigation treatments and the procedure were the same as in 1919 and 1920. The irrigations given in 1921 were as follows: Rows 3 and 11 irrigated on June 7 and 8 with 8.80 acre-inches to the acre; rows 5 and 13 on August 8 with 10.50 inches; and rows 7 and 15 on October 7 with 10.00 inches.

TABLE 16

SUMMARY OF YIELDS FROM MUIR PEACH ORCHARD, DAVIS, 1921

Row	Number of trees	Dates irrigated	Total weight fresh fruit, including culls	Culls	Weight fresh fruit	Weight dried fruit	Number of fresh fruit to make 1 pound of dried fruit
			Pounds	*Pounds*	*Pounds*	*Pounds*	
1	6	No irrigation	1666.5	222.5	1444.0	211.5	6.8
9	6		960.5	140.0	820.5	194.0	4.2
	Average to a tree		218.9	30.2	188.7 ±18.23	33.8 ±2.69	
3	6	June 7–8	1331.0	230.5	1100.5	226.0	4.8
11	5		1177.5	162.0	1015.5	215.5	4.7
	Average to a tree		228.6	35.7	192.4 ±6.53	40.1 ±1.26	
5	6	Aug. 8	1571.0	172.5	1408.5	249.0	5.7
13	6		1742.5	279.0	1463.5	249.0	5.9
	Average to a tree		276.1	37.6	239.3 ±15.44	41.5 ±2.62	
7	6	Oct. 7	1457.0	162.5	1294.5	258.0	5.0
15	6		1372.0	223.5	1148.5	221.0	5.0
	Average to a tree		235.8	32.2	203.6 ±13.03	40.0 ±2.44	

During 1922 rows 3 and 11 were irrigated on June 23 with 8.00 inches, rows 5 and 13 on August 12 with 10.00 inches, and rows 7 and 15 on September 20 with 10.50 inches. The soil-moisture conditions in the different rows during the seasons of 1921 and 1922, were substantially the same as those given for the year 1919. For this reason they are not reported.

The yields for the season of 1921 are given in table 16. The first picking was on August 23, and the last on September 2. The yields

of green fruit from the different rows for the season of 1922 is given in table 17. The first picking was on August 26, and the last on September 5, 1922.

TABLE 17

SUMMARY OF YIELDS FROM MUIR PEACH ORCHARD, DAVIS, 1922

Row	Number of tree	Dates irrigated	Total weight fresh fruit	Culls	Weight fresh fruit	Average weight fresh fruit to a tree
			Pounds	*Pounds*	*Pounds*	*Pounds*
1	6	No irrigation	1561.5	348.5	1213.0	202.2
9	6		1087.0	107.0	980.0	163.3
Average			1324.2	227.8	1096.5	182.6 ±9.32
3	6	June 23	1458.8	238.5	1220.2	203.4
11	5		1349.5	160.0	1189.5	237.9
Average			1404.1	199.3	1204.9	219.1 ±9.92
5	6	Aug. 12	1681.0	342.0	1339.0	223.2
13	6		1826.0	216.0	1610.0	268.3
Average			1753.5	279.0	1474.5	245.1 ±12.59
7	6	Sept. 15	2294.0	149.5	1244.5	207.4
15	6		1613.5	306.0	1307.5	217.9
Average			1953.8	227.8	1276.0	212.7 ±6.62

ANALYSES OF PEACHES FOR SUGAR AND ACID CONTENT

Samples of fruit for analyses were taken from the different rows during the seasons of 1917, 1918, and 1919. Care was taken in selecting the samples to secure representative fruits of average ripeness and size. At each sampling, two peaches were taken from each tree, one from the south side, and one from the north side. Analyses were made of the samples from each tree. Samples were collected every other day, starting about 10 days before picking. The first samples were quite green, and the last were over-ripe. All were taken from the trees at 8:30 o'clock in the morning.

The results obtained showed no significant differences in percentage of soluble solids, most of which, presumably, would be sugars,

that could be attributed to the differences in irrigation treatments. A summary of the results of the determinations of soluble solids in the pulp and juice of the peaches for the season of 1918 is given in table 18. The results of acid determinations for the season of 1919 are given in table 19. The amounts of acid found in the juice likewise showed no significant differences. The variations in the rows receiving the same treatment were in many cases as great as the differences between the rows irrigated differently.

TABLE 18

SUMMARY OF PERCENTAGES OF SOLUBLE SOLIDS IN PULP AND JUICE OF PEACHES FROM MUIR PEACH ORCHARD, DAVIS, 1918

Row	Dates of irrigation	Dates samples were collected				
		Aug. 9	Aug. 12	Aug. 16	Aug. 22	Aug. 26
1	No irrigation	3.43	4.56	3.87	4.30	3.51
9		3.83	2.97	3.37	3.20	4.37
Average		3.63	3.76	3.62	3.75	3.94
3	May 21	4.45	3.99	.....	5.44	3.91
11	Aug. 12 Sept. 25	3.33	3.00	.....	3.30	3.53
Average		3.89	3.50	.....	4.37	3.72
5	Apr. 4	3.55	3.25	3.14	3.62	3.17
13	May 21 Aug. 12	3.73	3.63	3.23	3.17	3.23
Average		3.64	3.44	3.18	3.40	3.20
7	July 2	3.75	3.21	3.15	2.49	3.72
15	Sept. 25	3.78	3.06	3.47	3.21	2.99
Average		3.76	3.14	3.31	2.85	3.36
2		4.08	4.50	3.38	3.78	5.29
4	Feb. 4	3.78	3.73	2.88	4.21	3.33
6		3.55	3.39	2.83	2.68	2.82
8		4.07	3.02	3.19	2.84	3.82
10		3.28	4.61	3.39	3.82	2.76
12		3.66	3.61	2.77	2.93	4.49
14		3.83	2.55	2.50	4.04	3.92
Average		3.75	3.63	2.99	3.47	3.78

TABLE 19

ACID CONTENT OF THE JUICE OF PEACHES FROM MUIR PEACH ORCHARD, DAVIS, 1919, IN GRAMS TO A LITER OF JUICE

Row	Dates of irrigation	Dates samples were collected				
		Aug. 8	Aug. 11	Aug. 13	Aug. 15	Aug. 17
1	No irrigation	16.89	12.71	7.72	4.29	4.70
9		10.72	11.26	9.11	3.22	5.63
Average		13.30	11.98	8.41	3.75	5.16
3	June 5–6	11.69	8.58	7.72	4.07	6.03
11		11.79	9.38	11.39	3.75	3.75
Average		11.74	8.98	9.50	3.91	4.89
5	Aug. 4–5	8.58	10.72	7.51	4.29	5.63
13		10.72	13.71	9.87	4.51	4.02
Average		9.65	12.21	8.69	4.40	4.82
7	Sept. 11	9.65	11.53	8.26	4.07	5.63
15		11.80	11.26	8.26	5.90	4.02
Average		10.72	11.39	8.26	4.98	4.82

MEASUREMENTS OF GROWTH OF PEACH TREES UNDER DIFFERENT SOIL–MOISTURE CONDITIONS

Beginning with the season of 1919, measurements were made of the circumferences of the trunks just above the crown of the tree, and of the main branches of the trees near the point where they left the trunk. The measurements were taken each time at the same place.

The bark was carefully smoothed before each measurement by scraping away with a knife any rough edges which protruded beyond the general surfaces of the trunk or branches. Only in one or two instances were measurements recorded which seemed to be incorrect. The results obtained from the beginning of the season of 1919 to the beginning of that of 1922, calculated in square centimeters of area of cross-section of the trunks of the trees in the different rows, grouped according to the irrigation treatment they received, are given in table 20. The average areas of cross-section of the main branches of the trees for the different rows also were obtained for this same period. Since no significant differences were found in the measurements for the trees in the different rows, these data are not presented.

TABLE 20

AREAS OF CROSS-SECTION OF THE TRUNKS OF MUIR PEACH TREES, DAVIS
MEASUREMENTS IN SQUARE CENTIMETERS

Row	Number of trees	Season of 1919							
		April 25	May 15	June 6	June 14	July 3	July 29	Aug. 28	Sept. 18
1	6	221.6	226.0	230.4	230.2	234.6	238.2	243.4	247.0
9	6	182.5	186.0	189.1	188.2	190.9	192.3	193.8	195.2
	Average	202.1 ±4.69	206.0 ±4.83	209.8 ±5.37	209.2 ±5.32	212.8 ±5.31	215.3 ±5.69	218.6 ±5.95	221.1 ±6.26
3	6	200.2	203.8	206.6	207.3	211.7	215.0	219.6	223.4
11	5	182.1	185.3	187.8	188.3	191.7	199.4	201.2	204.6
	Average	191.2 ±8.63	194.5 ±6.95	197.2 ±7.40	197.8 ±8.63	201.7 ±7.40	207.2 ±7.45	210.4 ±7.15	214.0 ±7.99
5	6	179.8	182.5	186.1	186.5	188.3	193.7	194.1	197.2
13	5	217.7	220.8	222.4	222.5	225.8	228.7	232.0	235.7
	Average	198.8 ±8.12	201.7 ±6.62	204.2 ±4.80	204.5 ±6.02	207.1 ±6.08	211.2 ±6.43	213.1 ±6.24	216.4 ±6.20
7	6	197.6	198.0	202.9	203.4	203.8	208.1	209.9	212.8
15	5	202.8	208.2	211.0	210.6	214.2	219.3	221.3	224.3
	Average	200.2 ±5.62	203.1 ±5.89	207.0 ±6.19	207.0 ±6.20	209.0 ±6.43	213.7 ±6.89	215.5 ±7.18	218.5 ±7.41

Row	Number of trees	Season of 1920							Season of 1921		1922
		Jan. 13	May 5	May 21	June 9	July 1	July 20	Aug. 6	Feb. 25	June 7	Jan. 26
1	6	251.6	256.8	259.1	261.4	266.2	269.7	271.5	285.2	296.9	306.5
9	6	197.2	203.0	204.1	204.7	206.5	207.6	207.5	212.3	222.2	228.9
	Average	224.4 ±6.46	229.9 ±6.63	231.6 ±6.84	233.1 ±7.06	236.3 ±7.43	238.6 ±7.79	239.5 ±7.94	248.8 ±9.19	259.7 ±9.85	267.7 ±10.34
3	6	225.1	231.2	231.4	233.5	236.2	239.2	241.4	245.8	259.4	264.7
11	5	208.7	215.7	216.9	219.9	223.9	228.6	231.1	231.4	246.8	257.8
	Average	216.9 ±7.93	223.5 ±8.13	224.1 ±8.08	226.7 ±8.09	230.0 ±8.05	233.9 ±8.20	236.3 ±8.19	238.6 ±8.37	253.1 ±8.78	261.2 ±9.30
5	6	199.4	205.5	206.5	207.7	210.0	213.1	216.1	220.4	232.4	239.2
13	5	238.9	247.3	249.9	251.1	254.9	258.4	259.7	271.1	285.4	289.9
	Average	219.1 ±6.17	226.4 ±6.40	228.2 ±6.55	229.4 ±5.60	232.4 ±6.89	235.8 ±7.07	237.9 ±7.69	245.7 ±8.31	258.9 ±8.48	264.5 ±8.68
7	6	214.3	221.8	222.7	224.4	227.7	228.8	229.6	234.7	246.5	255.7
15	5	226.9	233.9	235.0	239.9	240.6	243.6	246.2	254.3	265.0	274.9
	Average	220.1 ±7.59	227.9 ±7.84	228.8 ±8.93	232.1 ±7.72	234.2 ±8.00	236.2 ±8.26	237.9 ±8.38	244.5 ±8.71	255.7 ±9.22	265.3 ±10.29

The peach trees were too large to permit taking length growth measurements directly. Estimates were made of the seasonal growth each year. With the exception of the year 1920, during which the trees in row 9 apparently did not make as good growth as the other trees, no apparent differences could be detected. Such estimates on trees so large as those under observation, are of doubtful value. Tufts[50] found high coefficients of correlation between circumference of trunk and weight of top, and also between circumference of trunk and weight of root, on two-year-old peach trees. This author calls attention to the fact that circumference measurements made to determine growth "take into consideration only quantitative changes in the plant, and pay no regard whatever to qualitative changes. For this reason, circumference measurements lose much of their value as soon as the trees cease their purely vegetative growth and prepare for the production of blossoms and fruit."

The writer, together with Professor A. H. Hendrickson, of the Division of Pomology, made measurements of total length of new growth and circumferences of trunk, in an orchard of French prune trees adjoining the Muir peach orchard under discussion. Data were secured from 195 trees which were being studied to determine the variability before starting an irrigation experiment. The trees were nursery stock, French prunes on Myrobalan root, planted in the orchard in February, 1917.

Measurements were made at the end of the growing seasons of 1919 and 1920. The trees bloomed and bore a few prunes in 1920. The coefficient of correlation between length growth and area of cross-section of trunk in 1920 was 0.74 ± 0.026. In January, 1921, the coefficient of correlation between length growth and area of cross-section was 0.71 ± 0.025, and the coefficient of correlation between length growth and increase in area of cross-section was 0.68 ± 0.026. In each case there was strong positive correlation, indicating that a measure which gives the area of the cross-section of the trunk, gives a satisfactory indication of the amount of new growth being made by prune trees in an orchard for four seasons.

While measurements of circumferences of trunks may not be an exact measure of the growth of older producing trees, the fact that no significant differences were found in either the increase in area of cross-sections of trunks, or of main branches of the peach trees under observation, may indicate that there was no difference in growth due to different soil-moisture conditions. Figure 21 graphically illustrates the data given in table 20, and shows that no significant difference in rate of increase in the cross-sectional area of the trunks

under the different treatments occurred from April, 1919, to January, 1922.

A record of the growth made by trees in the different rows was obtained by a photographic method which will be described in detail elsewhere. Comparisons made between measurements taken from photographs made at different times show there was no significant difference in the growth of the trees in the different rows with the exception of the season of 1919–1920, an unusually dry season during which trees in the unirrigated rows were wilted for a long period.

Discussion of Results

These studies with peach trees lead to the same general conclusions reached in the studies with the Santa Clara Valley prune orchards. Differences could not be observed in trees growing on soil with high moisture content and those growing on soil with low moisture content until the soil-moisture supply had been reduced to a condition corresponding to the calculated wilting coefficient.

The yields from the non-irrigated trees in rows 1 and 9 in 1917, 1918, 1920, and 1922, were less than those from the irrigated trees. The soil-moisture content in each of these years was reduced to the wilting coefficient early in the season and the trees remained in a wilted condition for a long time. There appear to be no significant differences in yields from the trees in the rows under the different irrigation treatments.

The size of the peaches did not seem to be affected by irrigation near the time of picking. Likewise, the quality as measured by the amount of water in the fruit was not influenced by a high moisture content in the upper 6 feet of soil near the time of ripening. Furthermore, the results of the determinations of the amount of soluble solids, most of which were sugars, and the acid content, though based on so few samples from each tree, also indicate that the quality of the fruit was not materially influenced.

As in the case with the mature prune trees, it was noted that these peach trees always wilted when the moisture supply of the first 6 feet of soil had been reduced to the wilting coefficient even though the soil below this depth was moist. It appears, then, that the number of roots below the 6-foot depth was not sufficient to keep the trees from wilting. However, except during the season of 1920 when the trees in the unirrigated rows were badly wilted and largely defoliated toward the end of the season, some turgid leaves would be found on the wilted trees even though the upper 6 feet of soil were dry.

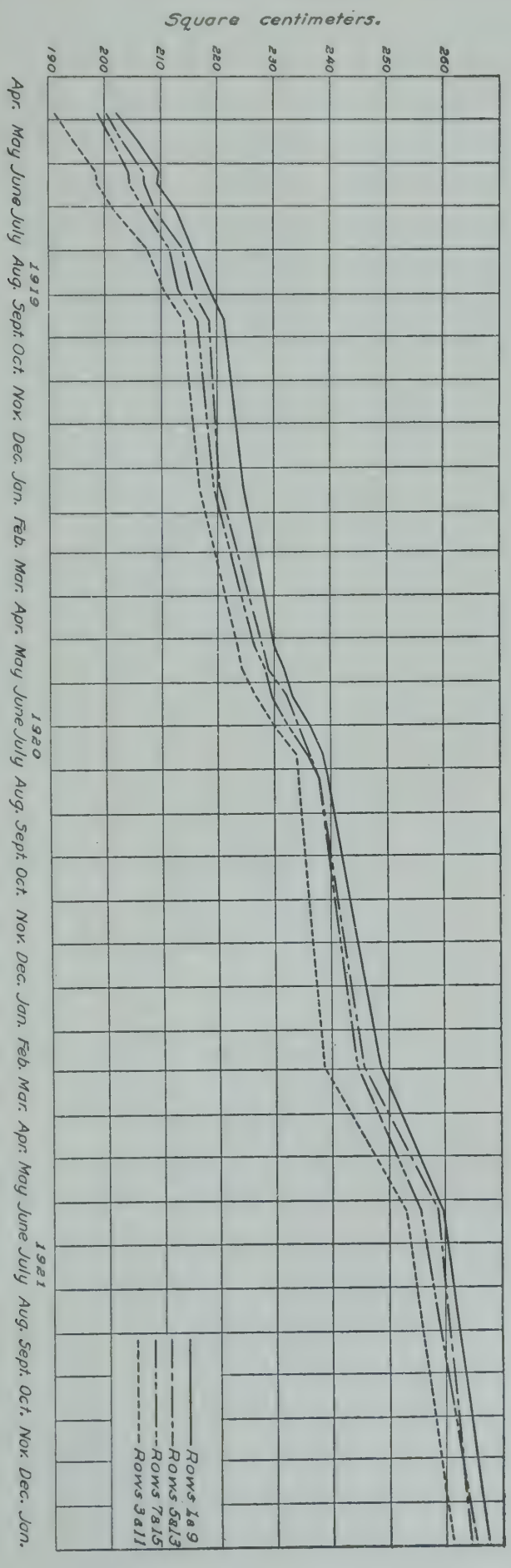


Fig. 20. Areas of cross-section of trunks of Muir peach trees, Davis.

The moisture content of the soil was not reduced to the same relative degree of dryness by these peach trees as by the mature prune trees. The lowest moisture content found in any of the plots in the Muir peach orchard was 7.5 per cent in August, 1920. The wilting coefficient of the soil in this plot is 8.8 per cent and the hygroscopic coefficient is 7.5 per cent.

The range of soil-moisture content for the soil of the plots in the Muir peach orchard between the maximum field or capillary capacity and the wilting coefficient is approximately 10 per cent. This is equivalent to about 9.5 acre-inches of water in 6 feet of soil. The fluctuations of water supply between these limits in this depth of soil did not seem to influence the growth of the trees.

The set of fruit buds did not seem to be influenced by the application in the fall of sufficient water to raise the upper 6 feet of soil to its maximum capillary capacity.

The use of water by trees and the effect of the maintenance of the soil-moisture supply between different ranges is discussed in the following section.

SECTION III

STUDIES OF TREES GROWN IN CONTAINERS UNDER CONTROLLED SOIL-MOISTURE CONDITIONS

The need for more definite information concerning the effect of different soil-moisture conditions on deciduous fruit trees and the use of water by these trees under these different conditions is apparent when an interpretation of the results obtained in the Santa Clara Valley and Muir peach orchard studies reported in the two foregoing sections is attempted. For this reason the studies* reported in this section were undertaken. Studies of plants grown in tanks or potometers have been numerous, and the number of papers dealing with this subject is large. The work of Briggs and Shantz,[13] Kisselbach,[36] and Fowler and Lipman,[27] are typical with respect to methods and equipment used. The studies reported herein are similar to these.

Trees were grown in large galvanized-iron tanks varing in size from 23.5 to 27.08 inches in diameter, and from 4 to 6 feet deep, holding from 1000 to 2000 pounds of moist soil. Each tank was protected with a tight fitting cover having a central opening for the trunk of the tree. The annular space between the trunk of the tree and the central opening in the cover was filled with absorbent cotton. The absorbent cotton extended a short distance up the trunk of the tree. A piece of oiled-cloth was tied around the absorbent cotton and trunk of the tree and sealed to the outside of the cover with an asphaltic roofing paint. The loss of water was thus confined almost entirely to that through the leaves, and rain water was entirely excluded.

Different kinds of deciduous fruit trees were grown in these tanks, but only the results obtained from the tests with French prune trees will be presented. The trees were standard nursery stock, grown on Myrobalan rootstock. They were from 3 to 4 feet high at the time of planting and were two years old.

* Professor A. H. Hendrickson of the Division of Pomology, University of California, has collaborated to a very substantial degree in the work on which this section is based. He has also very kindly read the manuscript and offered helpful suggestions, for all of which acknowledgment is gratefully extended.

Method of Packing the Soil in the Tanks and Planting the Trees

The soil was placed in the tanks in layers corresponding to those it occupied in place in the field, and was packed so that the volume weight or apparent specific gravity was the same as the volume weight of the undisturbed soil. The volume weight of the field soil at Davis was 82 pounds to a cubic foot, while that of the soil at Mountain View was 90 pounds to a cubic foot. The tanks were coated with cement mortar, leaving a very rough surface, in order to secure a closer contact of the soil with the sides of the tanks. The mortar was brushed on the inside of the tanks so that horizontal ridges were formed when the cement set. When the soil was packed in the tanks so treated, it was possible to add water to the surface of the soil in the tanks with little downward movement around the sides of the tank. It was found that an even distribution of the water applied could be secured in this way.

All of the tanks were provided with means to drain them. In most, especially the older tanks, a pet-cock was fitted to the bottom and a screen and layer of gravel about 1 inch in thickness were placed in the bottom before packing. The newer tanks, these 27.08 inches in diameter and 72 inches deep, were constructed with a chamber at the bottom with a pet-cock to collect any drainage water. However, it is of interest to note that only in one or two instances was water found collected at the bottom of the tanks. This indicates that the water used for the irrigations was not in excess of what the soils could hold. Samples taken after irrigation showed that the soil was wet throughout the full depth of the tanks.

The first lot of trees was planted in the tanks at Davis on April 15, 1919, and were grown there until October, 1920, when they were moved to the Deciduous Fruit Station of the University of California, at Mountain View, in the Santa Clara Valley. These trees were planted in soil which is classed as Yolo loam. The tanks had been filled with this soil in 1912, and alfalfa had been grown in the tanks for five years. Before the trees were set in the tanks, the alfalfa plants and the soil to a depth of about 10 inches were removed and the roots screened out of the soil. The soil was then replaced and tamped to the same compactness in all of the tanks. The weight of the empty tanks, and the weight of the water-free soil in each tank was known. The percentage of moisture in the soil could be calculated from the weights of the tanks at any time.

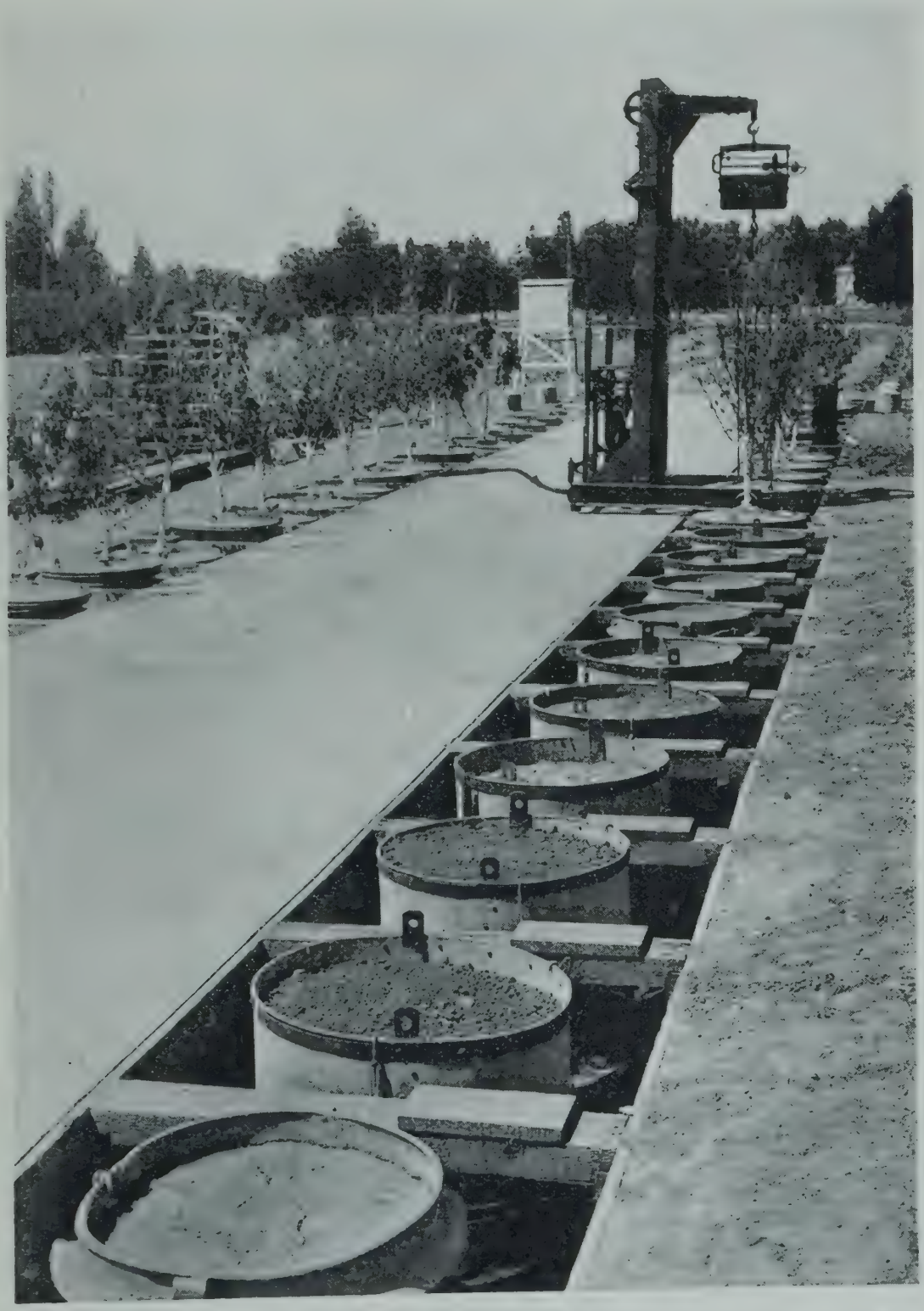


Fig. 21. Equipment for water-relation and evaporation studies installed at Davis.

The second lot of French prune trees was planted in the tanks at Mountain View on April 14, 1921. The soil used in these tanks was taken from a cherry orchard of about 25 years of age, the trees of which were growing vigorously, and which produced good crops in years when water was applied. This soil is classed as Yolo clay loam with gravel.

The soil adhering to the roots of the trees was washed away and the trees were pruned to whips about 20 inches high. The tanks were placed in trenches in order to prevent heating of the soil from exposure to the direct rays of the sun. To protect the tanks further from undue heating, the compartments of the trenches in which the tanks were placed were fitted with planks or sheathing cut to fit around the tanks. The equipment and its installation, after it had been moved to Davis in 1923, is illustrated in figure 21. The photograph was taken before the protecting sheathing was placed around all of the tanks. The trenches, which were arranged along the sides of an 8-foot concrete platform, were 3 feet wide and 4½ feet deep for the smaller tanks, and 3½ feet wide and 6½ feet deep for the larger tanks. The trenches were lined with 1-inch redwood planks nailed to framed bents or supports constructed of heavy timbers. The bents were spaced 41⅜ inches, center to center, which was the distance from one front wheel of the portable weighing derrick, to the other front wheel. Cleats were nailed to the tops of the bents, so that when the derrick was rolled onto them, the front wheels would stop at a position such that the hook on the weighing scales would be exactly over the center of the tanks.

Samples of soil in 1-foot depths were taken from each of the tanks containing the Yolo loam from Davis, and duplicate moisture equivalent determinations were made on each sample. Samples were taken at four places in each tank. The average moisture equivalent was found to be 22.0 per cent. Samples taken from the tanks containing the Yolo clay loam from Mountain View, also had an average moisture equivalent of 22.0 per cent. The Yolo clay loam samples, however, showed much greater variation in the value for the moisture equivalent than the samples of Yolo loam.

Method of Weighing the Tanks

The method of using the portable derrick is illustrated in figure 21. Two suspension scales were used throughout the tests, one for the smaller tanks weighing less than 1500 pounds, and one for the larger tanks weighing in excess of 2000 pounds. These scales were carefully

adjusted before any weighings were made, and checked against a master weight of 386 pounds. The scales gave satisfactory weights to within one pound. Weights of one pound placed on a tank while it was being weighed would be accurately recorded on the beam of the scales.

The portable derrick could be lengthened by the addition of an extension frame to permit tanks deeper than 6 feet to be moved from one compartment of the trench to another. Different sized weighing bales accommodated the derrick to the different sized trees to be weighed. However, in weighing, it was not necessary to lift the tanks out of the trench. It was only necessary to raise the tank a few inches until it swung clear of the bottom. The portable derrick with the scales and bales weighed almost a ton. However, it could be readily moved about the platform. It was possible to weigh 50 tanks within one hour with this apparatus. The weighings could be made on days with moderate winds which would prohibit weighings if the tanks had to be lifted out of the trenches. Most of the weighings were made early in the morning.

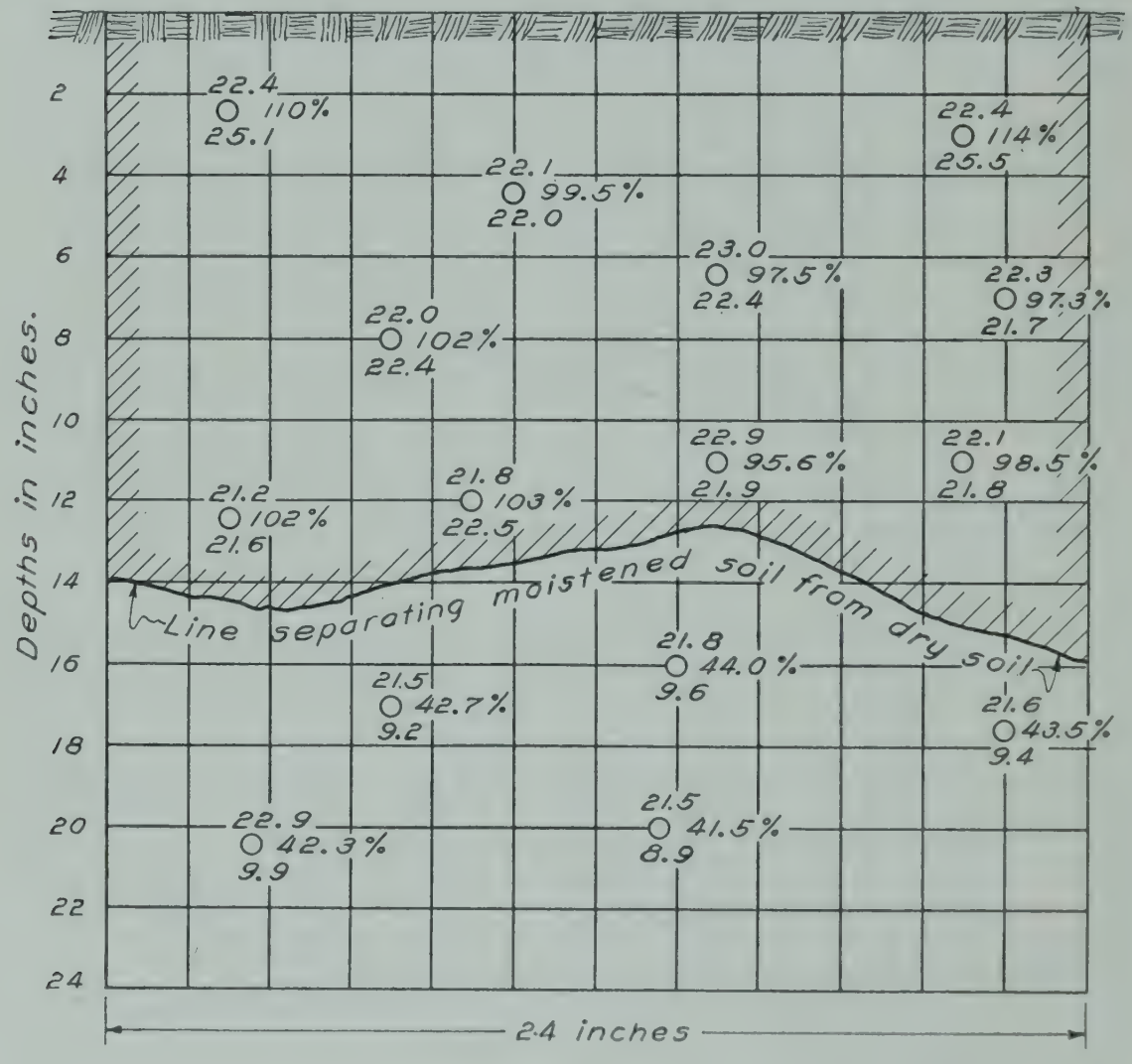
Distribution of Soil Moisture in the Tanks and Method of Applying Water

In the many experiments which have been made to determine the effect of variation in soil-moisture content upon plant growth, water has been added to the soil in different ways. Some have applied all of the water from above, others from below. In still other attempts, the applications have been made by means of specially arranged, perforated pipes. In the containers supposed to have a relatively low moisture content throughout the entire mass of soil there is evidence that the water applied was not uniformly distributed.

It is the belief of the writer that dependence upon capillary forces to bring about a uniform distribution in the soil of the water applied at any point has caused many erroneous conclusions to be drawn from water relation studies.* Attempts made during the present

* A paper (Shantz, H. L., Soil moisture in relation to the growth of crop plants, Jour. Am. Soc. Agron., **17**: 705–711. 1925), has appeared since this manuscript was prepared, in which the author expresses the same opinion, for he states: "Because of the peculiarities in the distribution of moisture in soils much of the work on the effect of varying water content on growth of crop plants, fungi, and bacteria, on the effect of varying water content on transpiration, water requirement, or the physical functions of the plant is entirely unreliable and will have to be repeated when conditions are known or better understood."

investigation to maintain a soil-moisture percentage less than that which the soil would hold against the force of gravity, the maximum field or capillary capacity, have met with failure in every case. During the course of these studies numerous trials were made, both with soils in tanks and with field plots, to maintain moisture contents less than the amounts of water the soils will hold against gravity, but it was found to be impossible to bring about relatively low moisture contents in the soil.



○ = Point from which soil samples were taken.
Upper numbers = Moisture equivalent determined by centrifuge method.
Lower numbers = Actuol moisture content of sample from field.
Percentages are the ratios of the moisture content to the moisture equivalent.

Fig. 22. Vertical distribution of water in a loam soil 48 hours after a rainfall of 2.15 inches. The upper margin of the diagram represents the soil surface.

An appreciation of the significance of this fact is vital to the proper interpretation of the results of water-relation studies with plants grown in containers, as well as in field plots. While the distribution of moisture by capillary action will be taken up in a later section, the following discussion may be helpful at this point. A picture of what actually takes place when water is applied to dry soils is given in figure 22. A trench was cut in a field of Yolo loam soil at Davis just after a rainfall of 2.15 inches. The field was level, and had produced a crop of barley the previous summer which had reduced the moisture content of the soil to an amount between 9 and 10 per cent. A system of coördinates was marked off on the face of the trench and samples were taken at the places indicated in figure 22. The line separating the moistened soil from the dry soil was sharply defined. The moisture content of the samples was determined, and the moisture equivalent determinations were then made on soil from these samples. The close agreement between the percentages of moisture, found in the moistened area, and the moisture equivalent is clearly shown in figure 22. There was an immediate drop in the ratio of the percentage of moisture to the moisture equivalent in the samples taken below the line of demarcation of wet and dry area. In all of the studies of the distribution of moisture after irrigation, this condition invariably has been observed. An application of a certain amount of water to a soil results in the wetting of that soil to its maximum field capacity to a definite depth, which depends upon the water holding capacity of the soil, and the initial moisture content.

Relatively small amounts of water applied to the surface of the soil in a tank in which a plant is growing will affect only the soil-moisture content to the depth of soil which can be raised to its maximum field capacity by this amount of water. The rate of extraction of moisture by the plant would usually be such that the moisture supply would be depleted long before additional downward movement could take place even if further capillary movement of moisture from the moist to the drier soil were appreciable in extent as some investigators assume. Alway and McDole[3] are two of the few investigators who have studied the downward penetration of definite amounts of water in soils and have considered the relative water retentiveness of the soils. These investigators found from the study of the downward movement of one inch of water applied to soils in glass cylinders about three inches in diameter that the applied water seemed to reach equilibrium at the end of five days in the finer-textured soils, but in the coarser ones it continued to move downward for a longer time.

However, the downward movement, in all cases, was relatively slow after the first hour, and in most cases, over 50 per cent of the movement which occurred in five days took place within the first hour after the water was applied. These investigators also pointed out that the moisture content of the moistened layer showed a rather constant relation to both the hygroscopic coefficient and the moisture equivalent.

In all of the soil-moisture studies made in connection with the work reported herein, such an agreement has been found immediately after irrigation, which in the loam soils used in these experiments was between 24 and 48 hours after irrigation, between the moisture content of the soil to the depth wetted and the moisture equivalent, that it is believed the latter can be used as a measure of the field capacity of these loam soils. Results of trials with the Yolo loam and the Yolo clay loam, used in the tanks, show that water in amounts sufficient to raise a certain depth of soil to a percentage equal to the moisture equivalent, resulted in wetting the soil to the desired depth. It is possible that there may be subsequent downward movement of moisture which continues for a long time and which results in an increase in moisture with increase in depth of soil when equilibrium is reached, as Israelsen and West[34] seem to show does occur, and which Gardner[28] thinks theory demands. However, in all of the tests made during these trials with the loam soils, there seemed to be a uniform distribution of moisture throughout the depth wetted. The extraction of moisture by plants on cropped soils was always sufficiently rapid to reduce the moisture content before further downward movement could be detected.

An amount of water was applied at each irrigation which would bring the moisture content of the soil to the desired depth up to the moisture equivalent or to the field capacity. It was found that in no case, either in the tanks with growing trees or in the field plots, could there be maintained a pre-determined moisture content less than the maximum field capacity of the soil. Therefore, the variation in soil-moisture conditions in these experiments was between the moisture equivalent, or the field capacity, and a certain minimum. For instance, a series of tanks was irrigated so that the soil-moisture content was maintained between 22 per cent, the field capacity, and 16 per cent. The tanks were irrigated when the weight of the tank indicated that the moisture content of the soil had been reduced to 16 per cent; others were irrigated when the moisture supply was reduced to the wilting coefficient, and so on. Therefore, instead of attempting to maintain a definite amount of moisture in the soil, the moisture content was allowed to fluctuate through a definite range.

Fig. 23. Prune tree on soil with a water table 2½ feet from the surface. Photographed in the fall.

The graphs of the soil-moisture conditions in the Santa Clara orchards and the Muir peach orchard at Davis show this to the the actual condition in a growing orchard. The moisture supply may be kept above any desired minimum, and that is all that can be accomplished.

The surface of the soil in the tanks was kept level, and all of the water was applied on the surface. The soil was kept in close contact with the sides of the tanks so that there would be no downward movement around the sides of the tank. In this manner a uniform depth of penetration was obtained. A slide in the cover could be removed to permit the water to be applied from a hose. The tanks were irrigated with tap-water from the domestic water supply. The amount of water added was weighed on the suspension scales and recorded in pounds.

In some of the tanks containing the trees, a water table was maintained at 2½ feet below the surface of the soil. This arrangement is illustrated in figure 23. An outer tank, serving as a reservoir, was supported around the inner tank which contained the growing tree. Water was added to the reservoir through the filling tube. A gage attached to a float was placed in the tube and the position of the gage indicated the position of the water table. The inner tank was perforated with many small holes to permit entry of the water. The annular space between the reservoir and the inner tank was covered with several thicknesses of oiled-cloth, which were securely sealed to both tanks by a coat of asphaltic roofing paint. This prevented the entrance of rain, and also prevented evaporation from the surface of the water in the reservoir.

Root Systems of Trees Grown in Tanks

Apparently Myrobalan root stock is well adapted to water-logged soils, since the trees growing in the tanks with a high water table showed no ill effects, appearing to be vigorous and healthy at all times. Prune trees on Myrobalan roots growing in water-logged soil in tanks for three seasons showed no injury. The root system of tree No. 1, which had been in soil with a water table 2½ feet from the surface, during the previous growing season, is illustrated in figure 24. The soil was washed from around the roots of the tree, and the tree was carefully removed from the tank. The soil was thoroughly permeated with roots and there was a matted mass of roots below the plane where the water had been maintained. The great mass of roots which grew below the water surface is clearly

shown in figue 24. At the time the tree was removed from the tank, these roots were making growth. The typical condition of the root systems of trees grown in soils in the tanks normally irrigated is shown in figure 25. This is a photograph of tree No. 22, for which data are presented in the following pages.

Fig. 24. Tree No. 1, French prune on Myrobalan root, grown with a water table 2½ feet from the surface. Lower matted mass of roots grew below the water surface. Trunk and branches weighed 3¾ pounds, roots 4½ pounds. Tree planted in tank April 15, 1919; photographed January 31, 1923.

The root systems of all of the trees except those in water-logged soil were found to be similar to that illustrated in figure 25. The soil was permeated with roots and the trees were pot-bound. Undoubtedly this condition influenced the growth of the trees and resulted in much less growth being made than under normal field conditions. However, since the tests were designed to indicate only the relative use of water and the response to different soil-moisture

conditions, this was not a disadvantage. The calculation of moisture present in the soil at different times usually was based upon the total weight of water-free soil in the tanks. Therefore, it was essential that all of the soil be penetrated by the roots of the trees at the time the tests were made. The calculated percentage of moisture was the average moisture content of the entire soil mass, and if there had been portions of the soil unoccupied by roots, or in which there was relatively few roots, these portions would have a higher moisture content than the calculated average. The moisture percentages calculated from the known weight of dry soil in the tanks were checked at times by taking soil samples from the tanks with a soil tube and the percentages of moisture obtained in these two ways usually agreed.

As mentioned before, it has been found during these studies that the capillary movement of moisture in these loam soils is very slow, and as the soil-moisture content decreases, the movement becomes practically negligible, resulting in little readjustment of moisture in the soil. The result is that the tree may not be responding entirely to a soil-moisture content calculated to be the average, if the soil in the container be not thoroughly permeated with roots. The pot-bound condition of the trees was therefore an advantage.

Weaver, Jean, and Crist,[52] studying the developments of the roots of plants grown in containers, also find that "the movement of water by capillary plays a rather unimportant role in replenishing water at the various levels from those adjacent." Briggs and Shantz[12] recognized uneven root distribution and lack of adjustment of the soil-moisture content through capillary movement as a source of error even in the very small containers in which they grew seedlings.

Climatological Measurements

Climatic conditions were measured by means of maximum and minimum thermometers, a hygrothermograph, and a hygrodeik exposed in a standard shelter about 4 feet above the ground surface, an anemometer set about 4 feet above the ground surface, a pyschrometer, a rain gage, and an evaporation tank. The evaporation tank was 3 feet in diameter and 3 feet deep, set in the ground 2.75 feet. The evaporation was read with a hook-gage to 0.01 inch. The evaporation tank, its construction and the manner of reading, were similar to the evaporation tanks and methods used by Sleight,[46] who showed the relative evaporation for tanks of various sizes and under

different conditions. Briggs and Shantz[14] point out that although the evaporation from a deep tank showed practically no correlation with transpiration when hourly values were considered, a discrepancy which results from the storage of heat energy in the large mass of water during the day, there is a correlation between the daily evaporation from a deep tank and daily transpiration.

Fig. 25. Tree No. 22, French prune on Myrobalan root, normally irrigated. Trunk and branches weighed 3½ pounds, roots 4½ pounds. Tree was planted in tank on April 15, 1919; photographed January 30, 1923.

Since the greater part of the experiments were conducted during 1922, some of the measurements of the climatic conditions at Mountain View for the growing season of this year, are summarized in table 21.

TABLE 21

SUMMARY OF CLIMATOLOGICAL MEASUREMENTS AT MOUNTAIN VIEW, DURING THE GROWING SEASON, 1922

Month	Days (inclusive)	Temperatures					Precipitation total for 5-day period	Evaporation total for 5-day period	Average wind velocity per hour
		Average of			Maximum	Minimum			
		Means	Maximums	Minimums					
		° *F.*	° *F.*	° *F.*	° *F.*	° *F.*	*Inches*	*Inches*	*Miles*
March	1– 5	47	59	35	64	31	.30	.36	2.8
	6–10	49	60	38	63	32	.14	.36	2.8
	11–15	48	58	37	63	31	.43	.20	2.2
	16–20	49	62	36	72	33	.25	.39	1.6
	21–25	52	64	41	67	36	.04	.26	1.8
	26–31	54	63	44	64	37	.19	.36	2.4
April	1– 5	54	67	40	75	38	.....	.55	2.4
	6–10	60	76	43	70	30	.....	.70	2.8
	11–15	43	59	37	62	33	.08	.59	3.5
	16–20	52	71	33	79	31	.....	.85	2.1
	21–25	53	67	39	74	36	.....	.66	1.7
	26–30	54	72	35	79	34	.....	.88	1.7
May	1– 5	62	83	41	89	40	.....	.89	1.6
	6–10	53	65	41	73	39	.28	.69	2.3
	11–15	66	89	44	96	36	.....	.89	2.3
	16–20	64	73	54	77	50	.05	.82	1.7
	21–25	56	73	40	79	38	.....	1.11	2.4
	26–31	60	78	43	89	36	.....	1.26	1.4
June	1– 5	59	72	46	74	41	.....	.81	1.4
	6–10	63	75	51	79	43	.03	.92	2.7
	11–15	63	74	52	85	45	.....	.97	2.5
	16–20	66	86	46	96	44	.....	1.21	1.3
	21–25	67	87	47	97	45	.....	.93	1.3
	26–30	66	76	56	89	54	.....	1.38	1.6
July	1– 5	68	80	57	86	50	.....	1.11	1.2
	6–10	66	77	55	80	48	.....	1.16	1.3
	11–15	68	80	55	92	50	.....	1.23	1.3
	16–20	66	79	52	84	50	.....	1.15	1.2
	21–25	60	73	46	75	41	.....	1.45	1.3
	26–31	64	80	48	85	45	.....	1.30	1.0

TABLE 21—(*Continued*)

Month	Days (inclusive)	Temperatures					Precipitation total for 5-day period	Evaporation total for 5-day period	Average wind velocity per hour
		Average of			Maximum	Minimum			
		Means	Maximums	Minimums					
		° *F.*	° *F.*	° *F.*	° *F.*	° *F.*	*Inches*	*Inches*	*Miles*
August	1– 5	64	76	52	80	48	.....	1.03	1.1
	6–10	62	79	46	90	44	.....	.97	1.1
	11–15	64	81	48	83	42	.....	1.13	2.4
	16–20	62	77	47	82	45	.....	1.05	1.5
	21–25	62	77	46	87	43	.....	1.02	1.1
	26–31	66	82	49	95	45	.....	1.26	1.1
September	1– 5	64	82	47	90	46	.....	.87	1.1
	6–10	70	92	49	100	51	.....	.83	1.0
	11–15	72	88	56	92	51	.....	.91	.7
	16–20	66	83	49	95	53	.....	.72	.6
	21–25	64	81	46	85	43	.....	.76	.8
	26–31	62	80	43	86	37	.....	.76	.8
October	1– 5	60	72	48	74	41	.20	.33	1.1
	6–10	61	74	48	79	44	.33	.36	1.2
	11–15	58	69	46	75	42	.....	.45	.7
	16–20	58	72	45	80	38	.....	.38	.5
	21–25	60	81	39	88	37	.....	.48	.4
	26–31	62	76	47	70	31	.43	.38	.9
November	1– 5	48	63	34	68	31	.....	.37	.9
	6–10	53	59	47	60	40	1.33	.....	1.2
	11–15	49	64	34	67	30	.....	.30	.4
	16–20	50	60	39	65	33	.....	.15	.3
	21–25	50	66	34	70	32	.....	.19	.2
	26–31	46	59	33	66	28	.....	.22	.6

Use of Water by Prune Trees in Soils with Varying Ranges of Moisture Content

The moisture treatment of the soil on which the trees had been growing up to the beginning of the season of 1922 had been substantially the same in all cases. In 1922, at Mountain View, the moisture content of some of the soils was allowed to fall below that of others, and the soil in some of the tanks was water-logged. A water table was constantly maintained at 2½ feet from the surface of the soil in the water-logged tanks. In addition to supplying water to the outer reservoir through the filling tube shown in figure 23, water was applied at frequent intervals during the season to the surface of the soil in these water-logged tanks.

Some of the trees were irrigated throughout the growing season when the moisture supply had been reduced to 16 per cent. Others were irrigated in this manner until the middle of August, and were then allowed to lower the soil-moisture content. In some cases, the soil-moisture content was allowed to fall to about the wilting coefficient before water was applied. Table 22 summarizes the use of water for the season of 1922 by some of the trees, grouped according to the irrigation treatment. The water transpired is assumed to be the total amount of water used by the plant. The amount of water retained in the plant and that used in its metabolism are so small in comparison with the amount of water transpired that they may be disregarded here.

The data in table 22 indicate the relative use of water by the prune trees in soils in which the range of moisture content varied. The use of water under high soil-moisture conditions can be compared with that under relatively low soil-moisture conditions. Such a comparison can best be made on the basis of use of water to a unit of leaf area. If there is a moisture condition optimum for growth or one above or below which the use of water by the trees is materially affected, it should be indicated by the use of water by trees under the different soil-moisture conditions maintained in these tanks. Obviously, a much more direct method would be to maintain definite moisture percentages in the soil rather than to allow the soil-moisture content to fluctuate. However, for the reasons previously stated this was found to be impossible.

TABLE 22

THE USE OF WATER IN POUNDS DURING THE GROWING SEASON OF 1922 BY FRENCH PRUNE TREES GROWN IN TANKS AT MOUNTAIN VIEW

Dates	Trees 3, 6, 9*‖			Tree 1†‖			Trees 12, 14†¶			Trees 5, 7‡¶			Trees 15, 17, 18‡¶			Trees 16, 19, 20§¶			Trees 4, 13, 21*¶		
	Per tree	Per tree per day	Per 1000 sq. in. leaf area per day	Per tree	Per tree per day	Per 1000 sq. in. leaf area per day	Per tree	Per tree per day	Per 1000 sq. in. leaf area per day	Per tree	Per tree per day	Per 1000 sq. in. leaf area per day	Per tree	Per tree per day	Per 1000 sq. in. leaf area per day	Per tree	Per tree per day	Per 1000 sq. in. leaf area per day	Per tree	Per tree per day	Per 1000 sq. in. leaf area per day
Mar. 1–15	2.6	0.2	0.06	1.0	0.1	0.03	5.0	0.3	0.23	3.5	0.2	0.10	1.0	0.1	0.06	2.0	0.1	0.03	3.0	0.2	0.07
Mar. 16–31	2.6	0.2	0.06	1.0	0.1	0.03	3.5	0.2	0.16	1.0	0.1	0.05	2.0	0.1	0.06	3.3	0.2	0.06	4.0	0.3	0.11
Apr. 1–15	5.3	0.4	0.12	4.0	0.3	0.09	5.5	0.4	0.31	1.5	0.1	0.05	3.3	0.2	0.13	4.3	0.3	0.09	3.3	0.2	0.07
Apr. 16–30	17.0	1.1	0.34	20.0	1.3	0.39	17.0	1.1	0.86	1.5	0.1	0.05	6.3	0.4	0.03	8.7	0.6	0.18	9.3	0.4	0.14
May 1–15	50.6	3.4	1.05	53.0	3.5	1.05	8.5	0.6	0.47	14.0	0.9	0.46	14.7	1.0	0.63	26.3	1.8	0.48	23.7	1.6	0.56
May 16–31	78.6	4.9	1.51	83.0	5.2	1.56	16.5	1.0	0.78	27.0	1.7	0.87	30.7	1.9	1.19	45.3	2.8	0.83	43.3	2.7	0.95
June 1–15	96.3	6.4	1.98	91.0	6.1	1.83	23.5	1.6	1.25	35.5	2.4	1.23	38.7	2.6	1.63	64.0	4.3	1.26	52.7	3.5	1.24
June 16–30	139.0	9.2	2.94	157.0	10.0	2.99	30.5	2.0	1.56	69.0	4.6	2.36	42.3	2.8	1.75	88.0	5.9	1.73	78.3	5.2	1.84
July 1–15	112.5	7.5	2.40	136.0	9.1	2.72	39.0	2.6	2.03	69.0	4.6	2.36	61.3	4.1	2.57	91.7	6.1	1.79	82.7	5.5	3.24
July 16–31	154.5	10.0	3.20	141.0	8.8	2.63	40.5	2.5	1.95	83.0	5.2	2.67	69.7	4.4	2.75	108.0	6.8	1.99	88.3	5.5	3.24
Aug. 1–15	105.5	7.0	2.24	143.0	9.6	2.87	45.5	3.0	2.42	83.0	5.6	2.82	56.0	3.7	2.32	75.0	5.0	1.47	67.7	4.5	1.59
Aug. 16–31	65.5	4.1	1.31	145.0	9.1	2.72	42.5	2.7	2.11	61.5	3.8	1.95	48.3	3.0	1.88	101.7	6.4	1.88	74.7	4.7	1.66
Sept. 1–15	106.0	7.1	2.27	143.0	9.6	2.87	47.0	3.1	2.42	74.5	5.0	2.57	32.3	3.5	2.19	110.0	7.3	2.14	65.7	4.2	1.48
Sept. 16–30	95.5	6.0	1.92	95.0	5.9	1.77	34.5	4.4	1.72	51.5	3.2	1.64	25.7	1.6	1.01	70.3	4.4	1.29	57.3	3.6	1.27
Oct. 1–15	37.0	2.5	0.80	48.0	3.2	0.96	22.0	1.5	1.17	24.0	1.6	0.82	15.3	1.0	0.63	46.7	3.1	0.91	29.0	2.0	0.71
Oct. 16–31	25.5	1.6	0.51	32.0	2.0	0.60	24.0	1.5	1.17	16.5	1.0	0.51	7.0	0.4	0.25	42.7	2.7	0.79	27.3	1.7	0.60

* Soil-moisture content ranged between maximum field capacity and wilting coefficient.
† Soil water-logged.
‡ Soil-moisture content maintained above 16 per cent until middle of August.
§ Soil-moisture content maintained above 16 per cent entire season.
‖ Planted in the tanks containing Yolo loam April 15, 1919.
¶ Planted in tanks containing Yolo clay loam April 14, 1921.

Trees, 3, 6, and 9, which were planted on April 15, 1919, in the tanks containing Yolo loam soil from Davis, were irrigated when the soil-moisture content was reduced to about the wilting coefficient. The tanks containing these trees were 23½ inches in diameter and 48 inches in depth. The trees were allowed to wilt a few times (see plate 1, figure 1). Tree 9 was irrigated when the soil-moisture content was reduced to the wilting coefficient, but on June 16, all of the leaves on the tree were removed in order to measure the loss of water from the bare limbs. The use of water by this tree after June 16 is not included in the average of trees 3, 6, and 9.

Tree 1, which was planted on April 15, 1919, in a tank 23½ inches in diameter by 48 inches in depth, containing Yolo loam, was grown in water-logged soil during 1922. A water-table was maintained at 2½ feet from the surface of the soil, and in addition water was applied to the surface. The use of water by tree 1 is listed in table 22 separately from that of trees 12 and 14, which were treated in the same manner as tree 1, but which were younger trees. Trees 12 and 14 were planted on April 14, 1921, in tanks containing Yolo clay loam. These tanks were 26 inches in diameter and 48 inches deep.

Trees 5 and 7 were planted on April 14, 1921, in tanks 26 inches in diameter and 48 inches in depth, containing Yolo clay loam. These trees were grown in soil kept above 16 per cent moisture until about the middle of August, when the soil-moisture content was allowed to fall to about the wilting coefficient.

Trees 15, 17, and 18 were of the same age and were treated in the same manner as trees 5 and 7. However, these were grown in larger tanks, the dimensions being 27.08 inches in diameter and 72 inches in depth. Since Kiesselbach[36] has suggested that the size of the tank may influence the water requirement of plants, the data for these trees are listed separately from those for trees 5 and 7. However, it should be noted that within the range of sizes of tanks used in these experiments, no differences in the use of water to the unit of leaf area were found. Trees 15, 17, and 18 were grown in soil with a moisture content above 16 per cent until about the middle of August. These trees were allowed to wilt several times. The wilted condition of trees 15 and 17 on October 18, 1922, is illustrated in plate 2, figure 1.

Trees 16, 19, and 20 were grown in soil which was kept above 16 per cent moisture content throughout the growing season of 1922. These trees were planted on April 14, 1921, in tanks 27.08 inches in diameter, and 72 inches in depth, containing Yolo clay loam.

Trees 4 and 13 were planted on April 14, 1921, in tanks 26 inches in diameter by 48 inches in depth, containing Yolo clay loam. Tree 21 was planted in the same soil on the same date but the tank was 27.08 inches in diameter and 72 inches in depth. These trees were irrigated when the moisture content fell to about the wilting coefficient.

Information concerning the dates on which some of these trees were allowed to wilt, and the soil-moisture percentage at the time of wilting, compared to the theoretical wilting coefficient is given in table 27.

The area of the leaves on the trees was obtained from the average measurements of several hundred leaves on different trees of the same age, and which were growing in soil maintained within the same range of moisture content. The average area of a leaf on the younger trees was 5.21 square inches; the average of the leaves on the older trees was 2.07 square inches. The total leaf area for each tree was found by multiplying the total number of leaves on each tree by this average area.

The use of water per unit of leaf area given in table 22 is not strictly correct for the spring and fall months. Few leaves were formed on the trees in the early spring, and many leaves dropped in the fall. The leaf area used as a basis for comparison is that measured about the middle of June. Measurements of leaf area before and after this period showed little change from May 15 to September 1. Furthermore, some of the trees were allowed to wilt after September 1 and consequently were partially defoliated. If it is assumed that the leaf area of the different trees in the fore part of the season was proportional to the maximum area measured in June, the data in table 22 for this period may be of value.

These data indicate that the use of water by these trees was not materially affected by the percentage of moisture in the soil. However, as will be presently shown, there was a very noticeable effect when the soil-moisture supply was reduced below the wilting coefficient.

It is assumed that the growing season of 1922 extended from March 1 to November 4, for on the latter date most of the leaves on the trees had matured or had wilted and dropped. The total use of water for the season of 1922 by the trees planted April 14, 1921, is given in table 23. Since some of the leaves on certain trees had dropped after September 25, the loss of moisture from March 1 to September 25 is listed, and is used in the calculations. The ratio of loss of water to leaf area, is recorded as loss in pounds to the square

inch of leaf area, and the ratio of the length growth made during the season of 1922 to the loss of water is recorded as the inches of growth for each pound of water used. The leaf area used for each tree was measured in June after length growth of the twigs had ceased.

The uniformity of the ratios of water loss to leaf area and new length growth is striking. The moisture content of the soil in which these trees were grown varied between wide ranges, yet the use of water by the trees was not materially affected. The use of water under similar atmospheric evaporating power seemed to be determined by leaf area. Tree 6, one of the older trees, showed a ratio of water loss to leaf area of 0.287 for the season of 1921, and in 1922 this ratio was 0.250, an agreement which is remarkably close.

TABLE 23

USE OF WATER BY YOUNG FRENCH PRUNE TREES GROWN IN TANKS AT MOUNTAIN VIEW. SEASON OF 1922

Number of tree	Length of growth in inches	Number of leaves	Leaf area in square inches	Water used per season, March 1 to Nov. 4, pounds to a tree	Water used from March 1 to Sept. 25, pounds to a tree	Loss of water to the square inch of leaf area	Inches of growth for each pound of water used
4*	536.0	756	3950	902	782	0.198	0.685
5†	348.0	394	2098	541	499	0.237	0.698
7†	403.5	346	1799	688	626	0.347	0.644
12†	214.0	239	1244	378	316	0.254	0.678
13†	363.25	474	2476	634	544	0.220	0.666
14†	221.0	253	1317	388	328	0.249	0.673
15‡	333.0	316	1644	464	427	0.259	0.778
16‡	365.0	510	2653	703	572	0.215	0.638
17‡	372.25	451	2347	647	587	0.250	0.634
18‡	185.0	154	803	302	261	0.325	0.709
19‡	491.0	627	3260	811	712	0.218	0.690
20‡	697.5	828	4305	1171	1020	0.237	0.683
21‡	351.0	398	2070	592	508	0.245	0.690
Average.....	375.42	442	2305	632	552	0.250 ±0.007	0.682 ±0.001

* Grown in tank 23½ inches in diameter and 48 inches in depth.
† Grown in tanks 26 inches in diameter and 48 inches in depth.
‡ Grown in tanks 27.08 inches in diameter and 72 inches in depth.

Coefficients of correlation were calculated from the data given in table 23 for the relation of use of water to leaf area and for the relation of use to length growth made during the current season.

The coefficient of correlation between water loss and leaf area is 0.97 ± 0.11. The coefficient of correlation between water loss and length growth is 0.995 ± 0.002. Coefficients of correlations with such high values as these, and so much greater than their probable errors, may be considered to be decidedly significant. It may, then, be safely said that the use of water by these young prune trees grown in clay loam soil in tanks, under the conditions prevalent at Mountain View, has not been materially influenced by the differences in amounts of water available for growth, and that optimum moisture conditions for growth cover a range of soil moisture from the maximum field or capillary capacity to about the wilting coefficient. Fowler and Lipman,[27] likewise, have suggested that the range of soil-moisture percentages within which young Lisbon lemon trees will grow satisfactorily in the loam soil studied by them is, relatively speaking, a wide one. However, the lower limit set by them is much higher than that observed in these studies. The maintenance of the lower percentages of soil moisture in the tanks used by these investigators may not have been uniform throughout the soil mass. The soil in the tanks containing the lemon trees which received the lesser amounts of water may have been wet to slight depths at each application of water instead of the entire soil mass being raised to the desired percentage of moisture. In this connection, it may be mentioned that, although Kiesselbach[36] recognized that the lack of uniformity in distribution of water applied to the soil in containers constitutes a serious source of error in experiments of this nature, it is not at all certain that the lower soil-moisture percentages that he attempted to maintain in his tanks were the true percentages of moisture throughout the entire mass of soil in the tanks. The moisture content of the soil in the immediate vicinity of his spiral irrigating coils undoubtedly was raised to the maximum field capacity and probably other portions of the soil were dry. Tests made at Davis by Professor S. H. Beckett from 1912 to 1917 with alfalfa growing in tanks to determine the use of water by these plants with varying soil-moisture contents proved that this was true in many cases. Water was applied to the tanks containing the alfalfa plants by means of perforated irrigating pipes arranged in spider-web fashion in the soil and also by means of spiral pipe irrigators. Attempts were made to maintain certain percentages of soil moisture in the different tanks calculated from the known weight of dry soil. Water was added to the soil to bring them up to the calculated weights. After the conclusion of the tests, some of the tanks were emptied and it was found that the roots of the plants grown in the soils presumed to be at the

lower moisture percentages were all within the soil in the immediate vicinity of the irrigating pipes. The soil in the bottom of these tanks probably was dry the greater portion of the time during the growing season. On the other hand, some of the tanks containing the soils assumed to be maintained at the higher moisture contents were found to be saturated at the bottoms.

Use of Water by a Young French Prune Tree Grown in a Tank Automatically Balanced

The individual behavior in the use of water of several of the prune trees grown in the tanks was studied closely. Where tanks containing 1000 pounds or more of soil were used, as in this case, no adequate weighing device capable of weighing less than one pound was available. Since it was desired to weigh very small losses of water, tree 22, growing in a tank 23½ inches in diameter by 48 inches in depth, containing Yolo loam, was automatically balanced, so that very small losses of moisture through transpiration could be recorded. The arrangement of the apparatus is illustrated in figure 26. The principle of operation of the device is essentially the same as the self-registering transpiration machine described by Copeland[22] and the auxanometer described later by Corbett.[23] However, it differs from Corbett's auxanometer in that the tank is counter-balanced. The tank containing the soil and tree was suspended in an outer tank which was set into the ground and contained water. The tank containing the tree was then balanced with a frame made of channel iron, carrying a weight at the outer end, and supported on a knife edge. As the tree lost water through transpiration, and thus lessened the weight of the suspended tank and contents, the tank containing the tree rose in the outer tank and a mass of water equal to that which had been transpired by the tree was replaced. The tank containing the tree automatically came to rest at this point, and a recording device attached to the outer end of this channel-iron frame, recorded the loss of the weight of the tree tank. The tank containing the tree was uniform in diameter at the section which moved through the water surface. The accuracy of the results would, of course, depend upon the uniformity of this cross-section of the tank.

Some difficulty was encountered during the first two years the apparatus was used, because the water would cling to the walls of the inner tank as it raised. However, in 1921 the inner and outer

tanks were coated with a patented varnish.* This varnish practically eliminated the adhesion of the water on the metal of the tanks.

The apparatus was run for two seasons at Davis, 1919 and 1920, and was removed to Mountain View and set up early in 1921. Measurements were taken frequently during these three seasons to determine shrinkage in the volume of water in the outer tank. The loss, for an entire season, by evaporation from the water in the outer tank was so small it could not be detected. There was very little fluctuation in the temperature of the water in the outer tank during the growing season, and consequently, it is probable that there was little change in the buoyancy of the water due to change in its density.

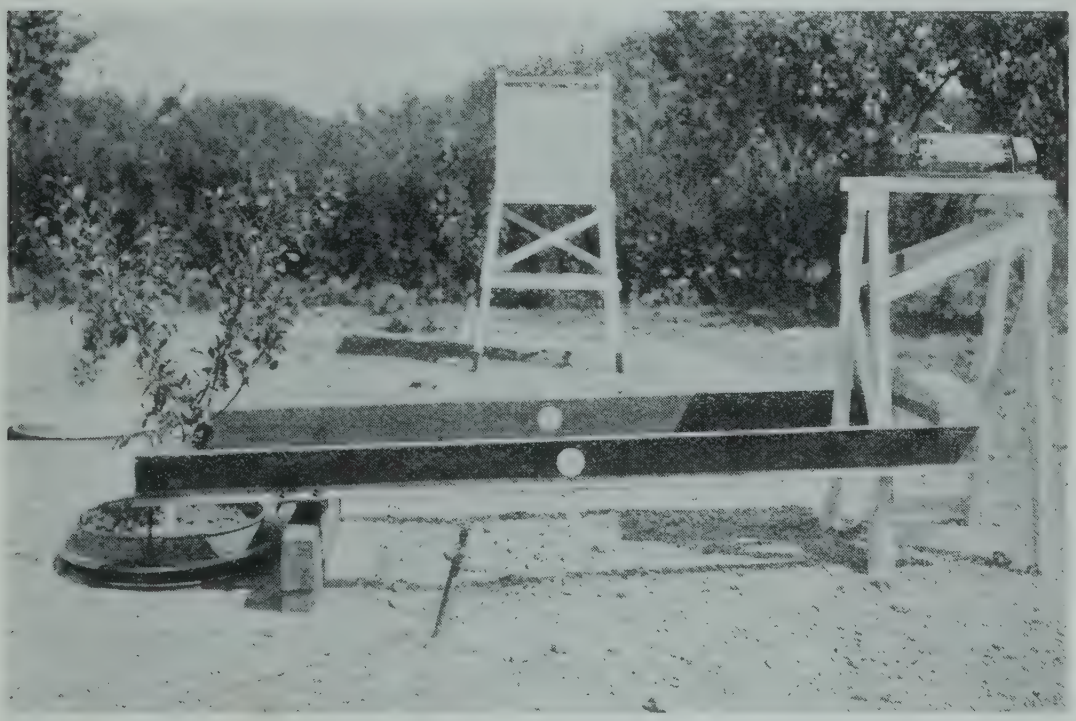


Fig. 26. Tree 22, in a tank automatically balanced so that small losses of moisture by transpiration could be measured. This photograph was taken in May, 1921, just after the apparatus had been moved from Davis to Mountain View. Later the tank was fitted with a cover and a wider rim and a low protecting wall was built around the apparatus to protect it from winds.

The movement of the outer end of the channel-iron frame was still further magnified in the recording device. The movement of the pointer on the record sheet was about 100 times the movement of the tank. The recording device was designed by Mr. E. J. Hoff, of the Division of Agricultural Engineering, Bureau of Public Roads, U. S. Department of Agriculture, and is essentially the same as the sensitive water-level recorder described by him.[32] The feature of the

* Made by the Gumite Corporation of New York City.

recorder lay in the elimination of friction, and in bringing the recording pen into contact with the record sheet every 30 seconds. This arrangement eliminated the resistance of the pen on the record sheet and allowed the recording arm to move freely and to respond to the movement of the tank. The apparatus illustrated in figure 26 was modified somewhat before starting the experiment in 1922. The recording device shown was changed to give a daily record instead of a continuous record. The tank containing the tree was covered and a wider rim was place around it to prevent the entrance of rain. Walls about two feet high were built around the apparatus to lessen the movement caused by winds. During the season of 1921, careful observations and repeated tests showed that losses of moisture by transpiration as small as 4 ounces could be weighed with confidence. In fact, the placing of 2-ounce weights on the tanks made a measurable movement of the pen on the record sheet. The apparatus was calibrated a number of times by placing standard weights on the tank containing the tree and noting the movement of the pen of the recorder. It was found that there was a definite movement of the pen for a definite weight. One pound of water lost by transpiration equalled six divisions on the record sheet. Three typical daily record sheets are shown in figure 27. The irregular lines in the portions of the graphs between about 10 A.M. and 7 P.M. are due to the movement of the tank caused by wind. The slope of the graphs indicates the rate of loss of water through transpiration. The recording drum revolved twice in 24 hours, and the recording sheet usually had to be replaced each day.

The tank was lowered and the channel-iron frame removed when the tank was irrigated. The tree tank was then raised clear of the water in the outer tank by means of the portable weighing derrick, and the desired weight of water was applied to the soil. The transpiration losses, recorded on the sheets, checked with the losses determined by weighing the tank with the suspension scales.

The data, taken from the record sheet of the automatic balance, showing the use of water by tree 22, summarized for 50 per cent of the time before each irrigation and 50 per cent of the time after each irrigation, is given in table 24. For instance, the tree was irrigated on June 3 and June 16; therefore, the average use of water is given from June 4 to June 9, and from June 10 to June 15. For the purpose of comparison, the average use of water in the forenoon and in the afternoon is given separately from the total daily use. The total number of degrees above 40° F. for each 5-hour period is also given

in order to indicate the relative amount of heat during the different periods. The climatological measurements taken at Mountain View during these periods are also summarized. At each irrigation the soil in the tank was raised to its full field capacity, 22 per cent, and in the majority of cases, water was not again applied until the

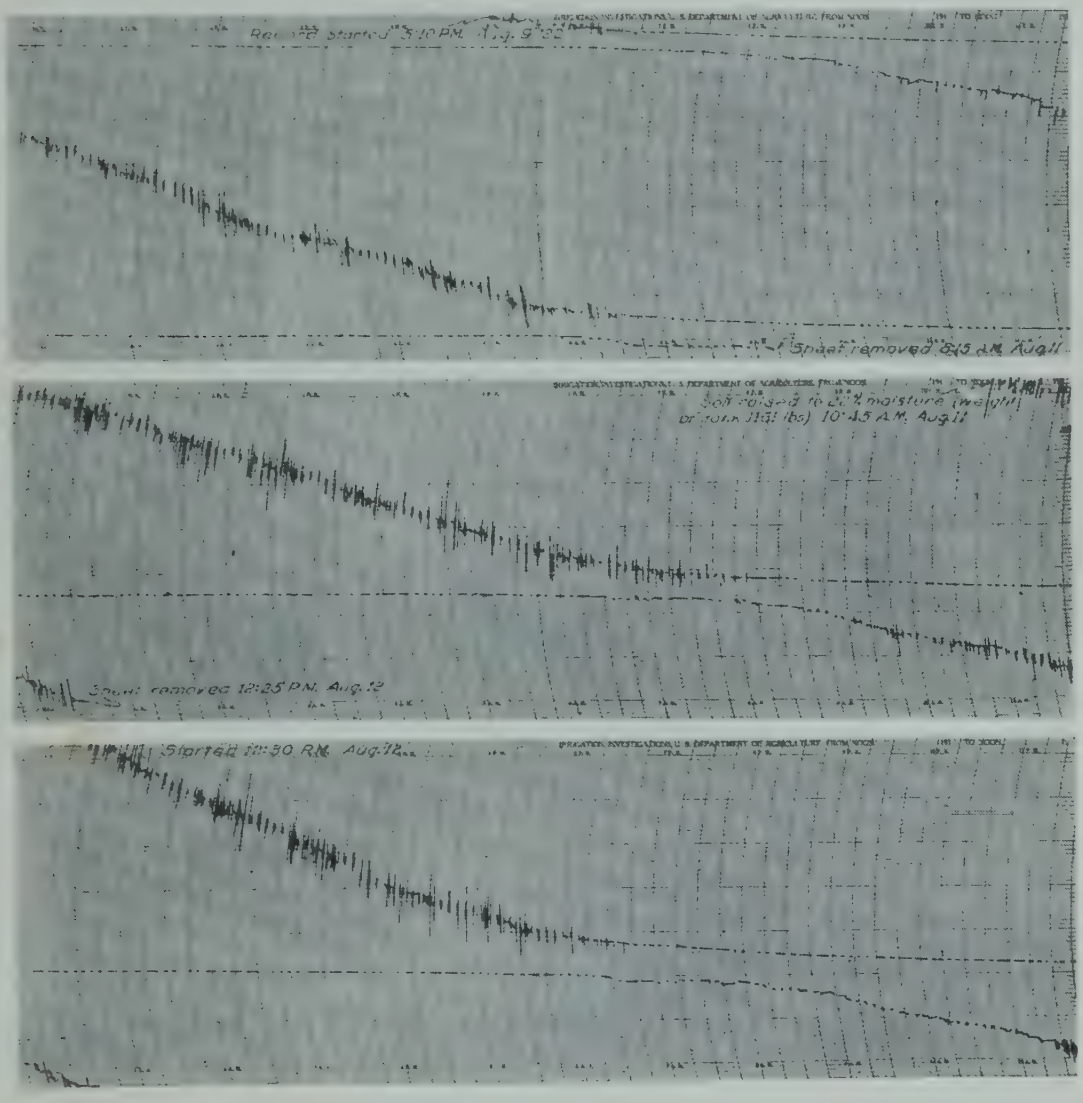


Fig. 27. Three typical daily record sheets taken from recorder used with balanced tank. The upper sheet is the record of transpiration the day before the water was applied, the middle sheet refers to that day, and the bottom sheet to the day following the application of water.

moisture content had been reduced to about 11.9 per cent, the wilting coefficient. Therefore, from the data in table 24, a comparison can be made between the rates of transpiration before and after each irrigation, when the soil-moisture content is high, and when it has been reduced to just above the wilting coefficient.

TABLE 24

Use of Water for the Season of 1922 by Young French Prune Tree 22 Growing in a Tank at Mountain View, Together with Climatological Measurements

Dates between irrigations for which data is summarized	Dates irrigated‡	Average use of water in pounds			Average evaporation per day	Average maximum temperature	Average minimum temperature	Average vapor pressure deficits, mm. Hg.†	Average wind velocity, miles per hour	Average total number of degrees of temperature above 40° F.*		
		8 a.m. to 1 p.m.	1 p.m. to 6 p.m.	Per day of 24 hours						8 a.m. to 1 p.m.	1 p.m. to 6 p.m.	8 a.m. to 6 p.m.
					Inches	° *F.*	° *F.*					
Apr. 25 to May 3	May 4	.99	.34	1.68	.17	75	38	8.94	1.6	145	159	304
May 5 to May 12		1.16	.57	2.22	.15	71	41	7.44	2.4	125	135	260
May 13 to May 19	May 20	2.15	1.65	4.94	.18	81	52	7.36	1.6	166	175	341
May 21 to May 26		2.68	2.41	6.28	.22	74	40	12.31	2.4	142	159	301
May 27 to June 2	June 3	2.37	2.26	5.78	.18	75	45	7.36	1.4	141	162	303
June 4 to June 9		2.35	3.24	6.49	.19	75	52	7.03	1.8	154	158	312
June 10 to June 15	June 16	2.23	2.86	6.84	.18	74	51	4.82	2.3	133	158	291
June 17 to June 19		3.00	3.94	8.47	.24	87	47	10.70	1.4	187	212	399
June 20 to June 23	June 24	5.10	4.36	11.27	.25	82	46	12.37	1.8	188	197	385
June 29 to July 3	June 28	4.35	4.70	10.45	.22	78	55	8.74	1.3	174	198	372
July 4 to July 7	July 8	4.08	4.26	9.27	.23	75	59	7.56	1.3	169	183	352
July 9 to July 12		4.31	4.67	10.50	.24	76	53	8.86	1.4	160	182	342
July 13 to July 16	July 17	4.60	4.12	10.15	.24	84	51	9.50	1.2	191	204	395
July 18 to July 21		4.54	3.84	10.15	.25	76	53	6.31	1.2	168	168	336
July 22 to July 24	July 25	4.72	3.98	10.06	.24	74	44	8.81	3.1	158	159	317
July 26 to July 28		3.83	4.74	9.99	.23	82	46	9.07	1.0	171	175	346
July 29 to Aug. 1	Aug. 2	3.56	4.08	8.93	.21	79	51	6.70	1.1	160	176	336
Aug. 3 to Aug. 6		3.72	3.66	8.79	.22	78	50	7.35	1.2	153	176	329
Aug. 7 to Aug. 10	Aug. 11	4.54	4.23	10.33	.19	76	46	8.44	1.1	153	176	329
Aug. 12 to Aug. 15		4.67	5.54	11.06	.25	82	46	14.12	2.4	166	194	360
Aug. 16 to Aug. 20	Aug. 21	4.10	4.70	9.83	.21	77	47	7.89	1.5	155	170	325
Aug. 22 to Aug. 25		4.14	5.18	10.58	.20	80	45	9.34	1.0	158	193	351
Aug. 26 to Aug. 29	Aug. 30	4.97	5.00	10.60	.20	81	51	9.62	1.1	172	188	360
Aug. 31 to Sept. 3		5.08	5.81	12.12	.20	88	46	9.41	.9	194	208	402
Sept. 4 to Sept. 7	Sept. 8	5.02	5.00	11.20	.18	82	46	13.39	1.3	165	196	361
Sept. 9 to Sept. 12		3.77	4.31	8.39	.16	92	55	10.50	.8	193	227	420
Sept. 13 to Sept. 17	Sept. 18	4.20	4.43	9.63	.18	88	52	10.71	.7	182	211	393
Sept. 19 to Sept. 23		2.80	3.03	6.87	.15	79	47	6.78	.6	142	170	312
Sept. 24 to Sept. 28	Sept. 29	3.73	3.53	6.70	.15	80	47	7.39	1.0	157	171	328
Sept. 30 to Oct. 7		2.40	2.35	5.78	.13	75	47	5.66	1.1	137	148	285
Oct. 8 to Oct. 15	Oct. 16	1.88	1.76	3.94	.09	70	48	3.49	1.0	107	127	234
Oct. 17 to Oct. 29		2.02	1.75	4.35	.08	73	41	5.84	.7	112	140	252
Oct. 30 to Nov. 12	Nov. 13	.95	.76	2.54	.07	62	39	3.03	1.0	81	79	160

* The number of degrees above 40° F for each hour was taken from the thermograph sheets and the total recorded for the 5 hours in the forenoon and the afternoon.

† Vapor pressure deficits measurements made at 5 p.m.

‡ At each irrigation the soil-moisture in the tank was raised to 22 per cent.

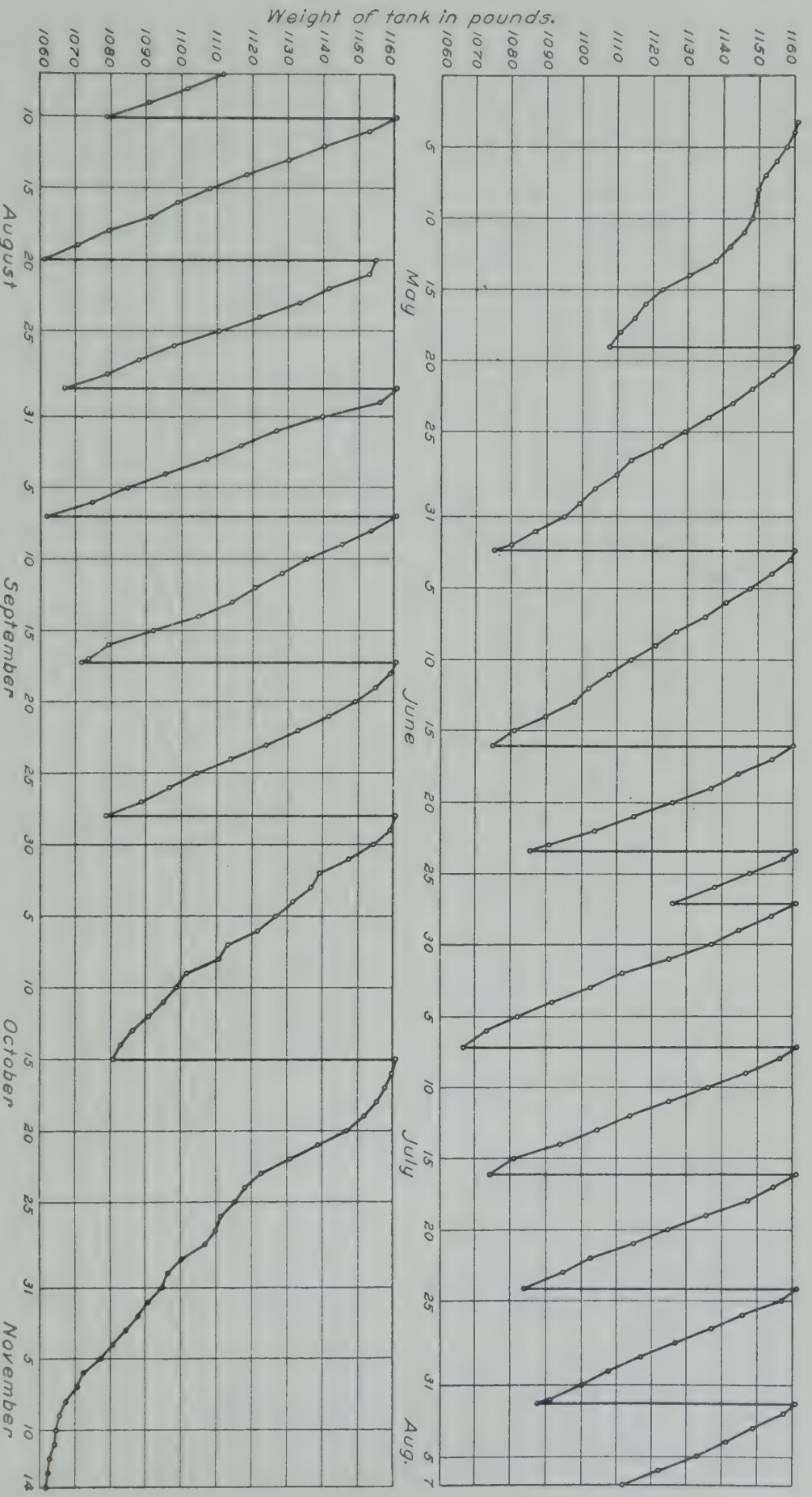


Fig. 28. Use of water by prune tree 22 during the season of 1922, at Mountain View. The weight of the tank with soil containing 22 per cent of moisture is 1161 pounds and the weight when the soil is at the wilting coefficient (11.9 per cent) is 1074 pounds.

The root system of tree 22, illustrated in figure 25, indicates that the soil in the tank was permeated with roots, therefore, the average moisture content of the soil, determined by weighing the tank and computing the moisture from the known weight of dry soil, probably gives a true value for the amount of water in contact with the roots of the tree. The data given in table 24 indicate that the use of water by this prune tree was no greater just after irrigation when the soil-moisture content was high, than it was when the moisture content was lower. The use of water both in the forenoon and in the afternoon before irrigation is about the same as that after irrigation. In some instances it is greater with low soil-moisture content than with high soil-moisture content and in others less. However, these differences are small and probably can be attributed to differences in the evaporating power of the air for the different periods. The agreement between the use with high and low soil-moisture content seems to hold throughout the entire growing season.

TABLE 25

SUMMARY OF THE MONTHLY USE OF WATER DURING THE SEASON OF 1922, BY YOUNG FRENCH PRUNE TREE 22

Month	Number of days for which record was obtained	Average use of water in pounds to the hour			
		8 a.m. to 1 p.m.	1 p.m. to 6 p.m.	For daylight hours	For 24-hour day
April	6	0.18	0.03	0.10	0.05
May	30	0.39	0.32	0.36	0.19
June	29	0.70	0.71	0.70	0.35
July	31	0.83	0.89	0.86	0.43
August	29	0.84	0.93	0.88	0.42
September	28	0.78	0.83	0.80	0.38
October	29	0.38	0.35	0.37	0.22
November	15	0.16	0.11	0.14	0.09

These facts are strikingly illustrated in figure 28, in which is plotted the daily use of water by tree 22. The weights of the tank, taken from the automatic record sheets on successive days, are plotted. The rate of use of water, which is indicated by the slope of the line representing the loss of moisture from the tank, is no greater immediately after irrigation when the soil-moisture content was 22 per cent than it is just before irrigation when the soil-moisture

content is about 11.9 per cent. When the climatological data, given in tables 21 and 24, are compared with the slope of the graphs in figure 34, it is apparent that conditions which made for high evaporation from a free water surface, likewise cause the tree to use greater amounts of water. Apparently, atmospheric evaporating power rather than stage of growth of the trees determines the use of water. Of course, the amount of evaporating surfaces exposed by the plant influences the use of water. Since there was practically no increase in leaf area after the middle of May, the increased use of water after this time can not be attributed to this cause. The records of the use of water by tree 22 are summarized for each month in table 25. The increased use in the warmer months is clearly indicated in this table.

Hourly weighings, by means of the portable device and scales, of other trees growing in tanks were made before and after irrigation several times during the season of 1922. In no case did there seem to be any difference in the amounts of water transpired before and after irrigation which could be attributed to the differences in soil-moisture content. The results from one of the trees which was irrigated on May 3 and on May 19, and weighed in this manner is given in table 26. These records were obtained from tree 6, which was planted on April 15, 1919, in a tank 23½ inches in diameter and 48 inches in depth, containing Yolo loam. Only the record of the weighings at 8 o'clock in the morning, at noon, and at 5 o'clock in the afternoon are recorded. Some of the climatological data are also recorded for comparison. There was no change in leaf area between May 3 and May 30. The low rate of transpiration from May 3 to May 9, which averaged only 2.6 pounds a day, compared to 5.3 pounds a day for the period May 10 to May 18, probably was due to differences in climatic conditions. The fact that the rate of transpiration between May 19 and May 30 was very uniform indicates that the transpiration was not measurably affected by the variation in amount of moisture above the wilting coefficient present in the soil.

It is interesting to note that this result was obtained in all of the trials made. The observations made after the removal of the equipment to Davis in 1923 also indicate that the use of water by the trees in these tanks was not influenced by the amount of soil moisture present, provided the moisture content was not reduced below the wilting coefficient.

TABLE 26

USE OF WATER AT MOUNTAIN VIEW BY PRUNE TREE 6

(Tree irrigated May 3 and May 19.)

1922	Weight of tank in pounds			Percentage of moisture in soil at 5 p.m. Calculated from weight of tank	Total loss of water through transpiration in 24 hours. *Pounds*	Evaporation from free-water surface in 24 hours. *Inches*	Wind velocity in miles to the hour	Total number of degrees temperature above 40° F.*
	8 a.m.	12 noon	5 p.m.					
May								
3	1050	1050	1120	21.7	.....	0.19	1.6	442
4	1119	1118	1116	21.3	4	0.20	1.7	314
5	1116	1115	1112	20.8	4	0.15	1.5	339
6	1112	1111	1109	20.5	3	0.18	1.3	277
7	1109	1108	1106	20.1	3	0.25	4.2	200
8	1105	1104	1103	19.7	3	0.19	2.5	186
9	1103	1103	1102	19.6	1	0.03	2.0	141
10	1101	1100	1098	19.2	4	0.04	0.8	228
11	1098	1096	1094	18.7	4	0.18	3.5	309
12	1093	1091	1089	18.1	5	0.19	3.6	401
13	1088	1085	1082	17.3	7	0.20	1.4	454
14	1080	1078	1073	16.2	9	0.12	1.4	502
15	1072	1069	1066	15.4	7	0.20	1.4	377
16	1064	1063	1060	14.7	6	0.19	1.2	313
17	1061	1061	1058	14.4	2	0.19	1.5	266
18	1056	1055	1054	14.0	4	0.20	1.8	225
19	1052	1050	1120	21.7	.....	0.17	2.7	248
20	1119	1118	1115	21.2	5	0.07	1.5	232
21	1113	1110	1107	20.6	8	0.24	2.4	293
22	1106	1104	1100	19.7	7	0.18	1.9	347
23	1099	1097	1094	18.7	6	0.23	1.9	285
24	1093	1091	1088	18.0	6	0.23	2.4	281
25	1087	1085	1081	17.1	7	0.26	3.3	232
26	1079	1076	1073	16.2	8	0.24	2.4	361
27	1071	1069	1066	15.4	7	0.21	0.8	442
28	1066	1065	1061	14.8	5	0.25	2.0	302
29	1061	1059	1055	14.1	6	0.18	1.0	341
30	1054	1053	1050	13.5	5	0.20	1.5	274

* The number of degrees above 40° F. for each hour taken from the thermograph sheets and the total is recorded.

The Percentage of Moisture in the Soil at the Time of Wilting of Prune Trees in Tanks

Throughout the several years of observation of the trees growing in the tanks, there were numerous opportunities to determine the moisture content of the soil when the trees wilted and would not revive until water was added to the soil. The dates on which the prune trees growing in tanks were allowed to wilt and the corresponding moisture content of the soil in which the trees were growing are listed in table 27. The trees were judged to be wilted when the leaves had lost turgidity and were not revived early in the morning, or until water was added to the soil. The nights at Mountain View were cool and generally the relative humidity was high, reaching 100 per cent at night many time during the growing season. Fogs frequently drifted in from the north from San Francisco Bay at night. At Davis, the nights were cool during the summer, but fogs were uncommon.

Briggs and Shantz[12] defined permanent wilting to be the condition at the time when the leaves of the plant first undergo a permanent reduction in their moisture content. By permanent reduction is meant a condition from which the leaves can not recover in an approximately saturated atmosphere without the addition of water to the soil.

Livingston and Koketsu[38] have further defined permanent wilting as that stage of progressive wilting from which recovery fails to occur within a period of 24 hours, if the wilted plants are exposed to a practically water-saturated atmosphere (in darkness) during that period. The large trees and containers used in these experiments made it impossible to place them in a moist chamber. Therefore, it is not known how far the condition of wilting (without recovery until water was added to the soil), adopted in these experiments, departs from the condition of permanent wilting as above defined.

The data presented in table 27 show close agreement between the percentage of moisture in the soil when these prune trees wilted without recovery until water was added to the soil and the calculated wilting coefficients for these loam soils. The wilting coefficients given are the averages of calculated values from duplicate moisture equivalent determinations made on two samples from each tank. All of the values have been averaged and recorded in the table. Weighed samples of 30 grams each were used in making the moisture equivalent determinations. When the weight of the tank at the time the

TABLE 27

Moisture Content of the Soil at the Time of Wilting of Prune Trees in Tanks, and Minimum Extraction of Moisture by the Trees

The equipment was moved to Davis in February, 1923. Observations and samples taken previous to this date were at Mountain View, and after at Davis.

Tree number‡	Date wilted	Weight of tank at wilting, pounds	Calculated weight of tank at wilting coefficient based on weight of dry soil in tank, pounds	Percentage of moisture in soil at wilting based on weight of dry soil in tank	Determined by centrifuge method*		Days after wilting before water was applied	Minimum weight of tank reached, pounds	Time in days to reach minimum	Percentage of moisture in soil calculated from dry weight of soil	Minimum moisture percentage reached, determined by sampling	Condition of tree
					Wilting coefficient	Hygroscopic coefficient						
3	May 3, 1922	1004	1000	11.6	11.1	7.5	4	998	4	10.9	.....	Tree revived.
3	Aug. 14, 1922	995	1000	10.5	11.1	7.5	8	982	6	8.9	8.1	Tree revived.
3	Sept. 28, 1922	1001	1000	11.2	11.1	7.5	33	980	24	8.0	8.0	Leaves yellow, finally defoliated.
4	Aug. 23, 1922	1032	1032	11.1	11.1	7.5	4	1011	4	8.6	8.5	Some yellowing, but tree revived.
5	Aug. 25, 1922	1356	1353	13.0	12.7	8.6	1	.....	.....	.....	.....	Tree revived.
5	Oct. 7, 1922	1353	1353	12.7	12.7	8.6	10	1343	3	11.8	11.3	Leaves yellow, finally defoliated.
5	May 3, 1923	1349	1353	12.3	12.7	8.6	3	.....	.....	.....	.....	Tree revived.
6	Aug. 21, 1922	1034	1037	11.6	11.9	8.1	1	.....	.....	.....	.....	Tree revived.
6	Sept. 2, 1922	1035	1037	11.7	11.9	8.1	1	.....	.....	.....	.....	Tree revived.
6	Apr. 29, 1922	1032	1037	11.3	11.9	8.1	4	1027	4	10.8	.....	Some yellowing but tree revived.
7	Aug. 22, 1922	1343	1355	11.0	12.1	8.2	1	.....	.....	.....	.....	Tree revived.
7	Oct. 7, 1922	1349	1355	11.5	12.1	8.2	10	1328	7	9.6	10.6	Completely defoliated.
9	Oct. 13, 1922	1074	1077	11.6	11.9	8.1	1	.....	.....	.....	.....	Tree revived.
10	July 7, 1924	1442	1437	11.9	11.6	7.9	1	.....	.....	.....	.....	Tree revived.
11	July 26, 1923	1318	1317	11.3	11.2	7.6	1	.....	.....	.....	.....	Tree revived.
11	Aug. 28, 1923	1320	1317	11.5	11.2	7.6	4	1309	4	10.0	.....	Some leaves yellowed and dropped.
13	June 12, 1923	1409	1411	12.1	12.3	8.3	1	.....	.....	.....	.....	Tree revived.
15	Sept. 14, 1922	1830	1837	11.0	11.5	7.8	61	1809	23	9.5	9.7	Leaves yellow, finally defoliated.
15	June 13, 1924	1912	1924†	11.0	11.8	8.0	1	.....	.....	.....	.....	Tree revived.
17	Aug. 28, 1922	1824	1830	11.8	12.2	8.3	1	.....	.....	.....	.....	Tree revived.
17	Oct. 7, 1922	1832	1830	12.3	12.2	8.3	38	1814	8	11.1	10.7	Leaves yellow, finally defoliated.
17	May 30, 1924	1900	1893†	12.3	11.9	8.1	3	1885	3	11.3	.....	Tree revived.
18	Aug. 12, 1922	1862	1865	10.6	10.9	7.5	7	1850	7	9.8	9.9	Some yellowing and defoliation.
18	Nov. 2, 1922	1863	1865	10.7	10.9	7.5	25	1862	2	10.6	10.5	Tree defoliated.
18	July 7, 1924	1896	1891†	11.5	11.2	7.6	1	.....	.....	.....	.....	Tree revived.
19	June 13, 1924	1941	1942	11.1	11.2	7.6	1	.....	.....	.....	.....	Tree revived.
20	June 13, 1924	1923	1929	11.0	11.4	7.8	1	.....	.....	.....	.....	Tree revived.

* Wilting coefficients calculated from moisture-equivalent determinations made on 30-gm. samples; 2 samples were taken from each tank and duplicate determinations made.

† Extra equipment and additional soil added to tanks in 1924.

‡ Trees 3, 6, and 9 were planted in Yolo loam, April 15, 1919, in tanks 23½ inches in diameter by 48 inches deep. Trees 15, 17, 18, 19 and 20 were planted April 14, 1921, in Yolo clay loam in tanks 27.08 inches in diameter by 72 inches deep. Trees 5, 7, 10, 11 and 13 were planted April 14, 1921, in Yolo clay loam in tanks 26 inches in diameter and 48 inches deep.

trees wilted and its weight at the theoretical wilting coefficient, calculated from the weight of dry soil in the tanks, are compared, the agreement is seen to be extremely close.

The equipment was moved to Davis in February, 1923. Therefore, observations and samples taken after this date were made at Davis and previous to this date at Mountain View. The climatic conditions which affect the rate of transpiration are markedly different in these two localities. Furthermore, the observations were made at different times during the growing seasons, so there were differences in atmospheric evaporating power, and yet the calculated wilting coefficient apparently is a measure of the moisture condition of these loam soils when the prune trees wilted and did not recover until water was added to the soil.

The results obtained by Briggs and Shantz[12] lead them to conclude that atmospheric environmental conditions have little or no effect upon the residual water content of the soil at the time of permanent wilting and that the wilting coefficient for any given soil is approximately constant for all species of plants grown on it and for all stages of their development. Caldwell[18] and Shive and Livingston,[47] who also studied the wilting of seedlings grown in small containers, found that the amount of residual moisture in the soil at the time of permanent wilting is dependent upon the intensity of atmospheric evaporating power for the period during which permanent wilting is attained.

Caldwell's[18] work seems to indicate that, while wilting under high atmospheric evaporating power always exhibited soil-moisture residue somewhat higher than the calculated wilting coefficient, this lack of agreement was less marked with the higher moisture holding capacity soils used by him. Mechanical analyses of the loam soils used for these prune trees show a silt and clay content of over 60 per cent, very fine sand about 20 per cent, and the remainder fine and medium sand. The soil used by Caldwell, which had the highest water-holding capacity, had a calculated moisture equivalent of 20.09 per cent, while the loam soils used in the present investigation had a moisture equivalent of about 22 per cent. Shive and Livingston[47] also show that the difference between the actual soil-moisture residue at permanent wilting and the calculated value become very small with the heavier soil of high water-holding power used by them.

The concurrence of the calculated wilting coefficient and the percentage of moisture in the soil at the time of wilting without recovery until water was added to the soil, has also been observed in the case of apricots and Calimyrna fig trees grown in tanks containing Yolo clay loam soil when studied in the same manner described in the

case of prune trees. However, fig trees have been more difficult to study under these conditions, since they drop their leaves more readily than the prunes or apricots when water is withheld. It is interesting to note that these three varieties of fruit trees apparently wilt when the moisture content of the loam soil on which they were growing was reduced to the same percentage at any time during the growing season.

The Minimum Reduction of Soil Moisture by Trees in Tanks

Some of the prune trees were allowed to remain for various periods of time in a wilted condition. These periods were generally toward the end of the growing season after other required data had been obtained from the trees earlier in the season. The minimum amounts of water found in the soil at the end of the time the trees were allowed to remain wilted are given in table 27. It should be noted that in no case was the water supply in the soil reduced to an amount equal to the calculated hygroscopic coefficient. Apparently, the trees in these tanks were only able to use about half of the water in the soil between the wilting coefficient and the hygroscopic coefficient. This is substantially the same result observed in the Muir peach orchard at Davis and in the Santa Clara Valley prune orchards, except in orchard No. 6 and orchard No. 2, where the soil-moisture supply was reduced to the hygroscopic coefficient.

The condition of trees 3, 4, and 5 on October 18, 1922, is shown in plate 1, figure 1. Tree 3 wilted on September 28, 1922, and tree 5 wilted on October 7, 1922. The soil-moisture content in the soil on which tree 4 was growing was kept above the wilting coefficient. The leaves on tree 3 were mostly dead and had completely collapsed, taking on a dark brown color before October 18. Tree 5 was decidedly wilted and the leaves were yellow and limp. Most of the leaves on tree 4 had turned yellow but all were turgid. This tree was maturing its leaves normally. The photograph, plate 1, figure 2, of the trees on October 28, shows the condition of trees 3 and 5 when the effects of drought had progressed still further. All of the leaves on tree 3 were black, and tree 5 was almost completely defoliated, while the leaves on tree 4 were dropping normally. A further example of the effects of withholding water for long periods of time is illustrated in plate 2, figure 1, by the condition of tree 15, which wilted on September 14, 1922. Tree 17 wilted on October 7, 1922; the soil in which tree 16 was growing was kept above the wilting coefficient. The photograph was taken on October 18, 1922.

The minimum reduction of the soil moisture by vetch grown in tanks was studied in the same manner as were the trees. The following may be cited as examples of these observations. On November 4, 1921, vetch seed was planted in a tank holding about 1000 pounds of soil. This tank was 23½ inches in diameter and 48 inches in depth. The vetch matured seed pods on June 1 and became permanently wilted. The weight of the tank on this date was 1121 pounds and apparently all loss of moisture from the tank had ceased by June 20, when the tank weighed 1116 pounds. The plants were removed and samples taken in foot depths to a depth of 3 feet. The moisture contents of these samples were determined and moisture equivalent determinations were then made. The following results were obtained:

Depth of soil sampled, feet	Percentage of moisture found in samples	Wilting coefficient	Hygroscopic coefficient
0 to 1	8.6	12.4	8.4
1 to 2	11.1	12.7	8.6
2 to 3	10.5	11.8	8.0

A tank, 26 inches in diameter by 48 inches deep, holding 1200 pounds of soil, was planted with vetch on the same date (November 4, 1921). More plants were allowed to grow in this than in the other. After the plants had matured and wilted and no further loss of moisture took place, samples were taken and the following results obtained:

Depth of soil sampled, feet	Percentages of moisture found in sample	Wilting coefficient	Hygroscopic coefficient
0 to 1	9.3	11.9	8.1
1 to 2	10.4	12.9	8.8
2 to 3	8.7	10.4	7.1

These records show that vetch plants and prune trees in these tanks used a considerable portion of the moisture in the soil below the wilting coefficient. Obviously the wilting coefficient is not the lowest limit of available water, although when the moisture content of these loam soils was reduced to a moisture content corresponding to the calculated wilting coefficient, the plants wilted and did not recover until water was added to the soil. These results agree with observations made in the Santa Clara Valley prune orchards and in

the mature Muir peach orchard at Davis. Therefore, it seems that the calculated wilting coefficient of the soils used in these experiments represents a critical soil-moisture condition. This soil-moisture constant is a better basis for comparing moisture conditions of soils than the hygroscopic coefficient, which probably represents the extreme lower limit of available moisture. Furthermore, the difficulty of directly determining the hygroscopic coefficient, as Puri[43] has shown, which also makes the value calculated from the moisture equivalent doubtful, mitigates against its use.

Effect of Soil-Moisture Content on Condition of Prune Trees in Tanks in the Fall

In connection with the study just described, observations were made of the condition of the prune trees in tanks in the fall under different soil-moisture contents. Trees 19 and 20 were irrigated so that the soil-moisture content was maintained between a range of maximum field capacity and 16 per cent throughout the entire season. The soil on which tree 18 was growing was kept above 16 per cent until about the end of the first week in August and then allowed to wilt and remain in this condition until August 12, 1922. Thereafter it was irrigated whenever the soil-moisture content had been reduced to about the wilting coefficient. The maximum field capacity of the soil in those tanks varied from 20 to 22 per cent. The wilting coefficient of the soil on which tree 18 was growing was 10.9 per cent; tree 19 was growing on soil with a wilting coefficient of 11.2 per cent, and tree 20 on soil with a wilting coefficient of 11.4 per cent. The condition of these three trees on October 18, 1922, is illustrated in plate 2, figure 2. More of the leaves on tree 19 had turned yellow than of those on trees 18 and 20, but the condition of these trees and leaves was substantially the same as that of trees with higher or lower soil-moisture content, provided those with lower soil moisture were not below the wilting coefficient. Trees which had grown on water-logged soil colored their leaves in the usual manner as early as did trees 18, 19, and 20. The condition of two of the trees, 13 and 14, which were on water-logged soil, is illustrated in plate 3, figure 1. The photograph was taken on October 18, 1922. Tree 13, the center tree in this figure, was on soil, the moisture content of which was just above the wilting coefficient. It was noted that many of the leaves of other trees on water-logged soil were yellow on October 18. The soil-moisture content for tree 13 had been maintained between the maximum field capacity and the wilting coefficient. Further illustra-

tion of the fall condition of trees on soils with different moisture contents is shown in plate 1, figure 1. Tree 3 was decidedly wilted, while tree 5 had been wilted for a short time before the photograph was taken. Tree 4, which was on soil with a moisture content just above the wilting coefficient, was holding its leaves normally, although many had turned yellow. Plate 1, figure 2, illustrates the condition of these trees 10 days later. The wilting of trees 3 and 5 had progressed further, while the leaves of tree 4 were normally dropping.

Defoliation caused by wilting could be induced by withholding water until the soil-moisture content was reduced below the wilting coefficient, but no differences in condition of the leaves or in the time the leaves dropped in the usual manner could be noted when trees on soil with high moisture content were compared with trees on soil with lower moisture content but not below the wilting coefficient.

This suggests that the time of beginning of coloring of the leaves in the fall and probably of dormancy or of maturity of the plant tissues is not affected by variations of soil-moisture content within a wide range. Furthermore, no injury from low temperatures during the dormant season could be detected in the trees in the Santa Clara Valley prune orchards and in the Muir peach orchard at Davis, which had been irrigated late in the season but before the leaves had dropped.

Use of Water During the Dormant Season by Prune Trees in Tanks

During the growing season, the pieces of oiled-cloth which had been tied around the trunks were loosened to allow them to expand. However, at the beginning of the dormant season, the pieces of oiled-cloth were sealed tightly around the trunks of the trees. Therefore, there was an opportunity to measure the losses of moisture, within the limits of accuracy of the weighing device, from the bare twigs and branches of the trees during the dormant season. The condition during the dormant season of three trees which had been growing in the tanks for three seasons is shown in plate 3, figure 2.

The results of the observations during the winter of 1922 at Mountain View are given in table 28. The losses are given for the period November 25, 1921, when all of the trees were completely defoliated, to March 1, 1922, the date on which the trees began to show activity again. For comparison, the losses also are given for the period, November 25, 1921, to April 1, 1922, at which time some leaves were formed on the trees.

TABLE 28

LOSSES OF MOISTURE BY EVAPORATION FROM THE BARE TWIGS AND BRANCHES OF PRUNE TREES GROWN IN TANKS DURING THE DORMANT SEASON OF 1922, AT MOUNTAIN VIEW

Number of of tree	Size of tank in which tree was growing	Date tree was planted	Loss of water Nov. 25, 1921 to March 1, 1922	Loss of water Nov. 25, 1921 to April 1, 1922
	Inches		*Pounds*	*Pounds*
1	23½ by 48	Apr. 15, 1919	1	2
3	23½ by 48	Apr. 15, 1919	3	8
4	23½ by 48	Apr. 14, 1921	3	7
5	26 by 48	Apr. 14, 1921	4	9
6	23½ by 48	Apr. 15, 1919	0	2
7	26 by 48	Apr. 14, 1921	2	6
9	23½ by 48	Apr. 15, 1919	4	11
10	26 by 48	Apr. 14, 1921	6	12
11	26 by 48	Apr. 14, 1921	8	5
12	26 by 48	Apr. 14, 1921	4	7
13	26 by 48	Apr. 14, 1921	8	15
14	26 by 48	Apr. 14, 1921	8	13
15	27.08 by 72	Apr. 14, 1921	5	9
16	27.08 by 72	Apr. 14, 1921	7	11
19	27.08 by 72	Apr. 14, 1921	2	8
20	27.08 by 72	Apr. 14, 1921	6	9
21	27.08 by 72	Apr. 14, 1921	5	.....
22	23½ by 48	Apr. 15, 1919	3	.....
Average			4.4 ±0.39	8.4 ±0.61

The soil in all of the tanks was fully wet on November 25, 1921, so that there was ample moisture available. While the covers fitted the tops of the tanks closely, they were not absolutely tight and, of course, some losses occurred by evaporation from the surfaces of the wet soil. No check tanks were available, consequently no estimate can be made of these losses. In this connection, it should be noted from the data in table 28 that apparently there is no agreement between size of the tree and the resulting loss. For instance, the large trees, such as trees 1, 3, and 6 shown in plate 3, figure 2, lost less than the smaller trees, such as trees 10, 11, 13, 14, and 15. A further example, tree 21, the tree shown in the left in plate 3, figure 2, lost five pounds, which was greater than the loss from any of the larger and older trees as the data in table 28 show.

A further example of the relatively slight losses of moisture through the young prune trees under observation when the leaves have been removed, is given in the results of the losses of moisture from tree 9, which was completely defoliated on June 16, 1922. This tree transpired 15 pounds of water during the 24 hours preceding the defoliation. From June 16 to July 1, the date on which the leaves began to open again, only 12 pounds of water were lost from the tank. On July 5, it was estimated that the tree was almost 3 per cent leafed out. Three pounds of water were lost between July 1 and July 5. On July 10 the tree was 20 per cent leafed out and had used 8 pounds of water between this date and July 5. During the week following July 10, the tank lost 40 pounds of water. The soil in the tank was wet to its maximum field capacity on the day the tree was defoliated. There were about 50 prunes on the tree, most of which stayed on until after the leaves were again formed.

The loss of water through these prune trees during the dormant season is relatively slight. Probably only in years of exceptionally light rainfall would prune trees in localities such as the Santa Clara Valley need irrigation during the dormant season so far as the current needs of the trees are concerned. However, as Batchelor and Reed[5, 6] have shown to be essential for walnuts, available water must be present during the dormant season.

Discussion of Results

During the course of the experiments described in this section, it was found impossible to bring about a uniform moisture content in the soil less than maximum field capacity. Applications of water to the surface of soils in containers or in field plots resulted in wetting the soil to a definite depth, depending upon the maximum field or capillary capacity and upon the initial moisture content. The water applied to the loam soils used in these containers raised the moisture content to the maximum field capacity in each tank, and this moisture content was established throughout the entire depth of soil penetrated by the water. The percentage to which the soil was raised seemed to be independent of the amount of moisture in the soil previous to the application of water. There seemed to be little subsequent movement of moisture and clearly there was no adjustment of moisture which would cause a uniform moisture condition to be established between the wet and drier soils.

It was found that the soil-moisture supply could be kept above a certain minimum. The soil could be raised to a maximum content and this condition reestablished when the plants had reduced the soil moisture supply to a certain minimum. All attempts to apply water, whether to the surface or from below, or by means of specially arranged, perforated irrigating pipes, in order to establish a uniform relatively low moisture content throughout the soil mass, met with failure. Experiments to determine the effect of varying moisture conditions upon the use of water by plants must be planned with these facts clearly in mind. Serious objections may be raised to previous water relation studies, wherein dependence has been placed upon capillary forces to bring about a uniform distribution in the soil of water applied at any point.

The data presented in this section show that the use of water by these prune trees grown in tanks was not affected by the total amount of water in the soil until the soil moisture supply was reduced to about the wilting coefficient. High coefficients of correlation were found between use of water and leaf area and the use of water and length of growth, even though the trees were grown on soils with varying ranges of moisture contents. These young prune trees, grown on clay loam soil in tanks under the conditions prevalent at Mountain View, were not influenced by the differences in amounts of water available for growth above the wilting coefficient. Optimum moisture conditions for growth cover a range of soil moisture from the maximum field capacity to about the wilting coefficient. In the loam soils used in these experiments there was a rather wide range of moisture content within which the trees seemed to thrive and develop equally well; it was equivalent on the average to a depth of 1.6 inches of water to a foot of soil. The moisture content corresponding to the wilting coefficient was a perceptibly dry condition, and one at which most orchardists would unhesitatingly judge the soil to be in need of water.

A number of investigators have shown that even though the moisture in the soil may fluctuate within a considerable range the plant will develop normally. However, the belief is prevalent that there is for each soil a particular water content, the optimum water content for growth at which plants grow best.

There appears to be no reason, either from physical considerations of the forces involved between the moisture and the soil particles or from physiological requirements of the plant, why optimum moisture conditions for growth should not vary from the maximum field capacity to about the wilting coefficient.

Shull[48] has shown that at the wilting coefficient, the soil withholds water from the plant with a force almost equal to four atmospheres, while the usual osmotic concentration of the sap of root cells of land plants is equal to seven or eight atmospheres. Under normal field conditions, therefore, the pressure gradient in favor of the plants should run from four to eight atmospheres, as the water holding power varies from zero to four atmospheres. Shull thinks that wilting at the wilting coefficient can not be due to lack of water, nor can it be due to equalization of forces between root hair and soil water. Therefore, he attributes the wilting of plants at the wilting coefficient to be due to the failure of water movement from soil particle to soil particle and from these to the root hairs, rather than from lack of moisture or of pressure gradient. However, Bouyoucos,[9] from the results of freezing point and dilatometer methods, believes that moisture near the wilting coefficient is held by the soil with such force that the plants can not extract it, and that the concentration of the soil solution is comparatively high, a factor which would tend to influence the intake of water by the roots. Bouyoucos holds that plants wilt, not because the soil moisture moves at an insufficiently rapid rate, but because it does not move at all. He points out that the dilatometer method results show that the percentage of moisture which fails to freeze at the supercooling of —1.5° C. is very nearly the same as that at which plants begin to wilt, a fact which indicates that the wilting coefficient of soils is at the point where the free moisture ends and the capillary adsorbed moisture begins. According to Bouyoucos' physiological classification of soil moisture, it is only the free water or the water which freezes at the supercooling of —1.5° C. that the plants can take up very readily, because very little force is needed to utilize it, since it exists in a free condition and is not held very rigidly by any outside force. The free or "the readily available" or "very available" water of Bouyoucos' classification corresponds to the moisture from the wilting coefficient to the field capacity. However, Bouyoucos[8] believes that the free water in the soil, the water between the wilting coefficient and the field capacity, would evaporate almost at the same rate as free water in mass, which indicates that he thinks the free water in the soil is held very loosely and very little if any force need be exerted to utilize all of it.

The rate of evaporation, vapor pressure, freezing point depression, wilting coefficient, hygroscopic coefficient, water holding capacity, and the unfree water content are all measures of the force with which water is held by the soil at different moisture contents. Parker[42] has discussed the different methods of determining the attractive force

the soil has for water and he shows that when the force with which water is held by the soil at different moisture contents is graphically represented the curves obtained by the rate of evaporation, vapor pressure measurements, freezing point depressions, and the method of Shull are all very similar. However, the curves differ in that the point of greatest curvature comes at different moisture contents. All of the curves show that for a wide range of moisture content there is only a slight increase in the force with which the water is held by the soil but the force increases very rapidly below a certain moisture content. Parker[42] points out that the freezing-point method and the method of Shull are more sensitive than the evaporation or vapor-pressure methods and that the point at which the force holding the water in the soil begins to increase rapidly will be at higher moisture content when measured by the freezing-point and probably by the method of Shull than the corresponding points obtained by the rate of evaporation and vapor pressure methods. However, all of the methods show that at low moisture contents, the water is held with much greater force than the additional water at the high moisture contents. Shull[48] shows a very slight increase in the force holding the water in the soil until the moisture content is reduced to the wilting coefficient, but below this the force increases very rapidly. The silt loam soil used by him had a wilting coefficient of 19.1 per cent. The increase in surface force from saturation to 18.87 per cent moisture was from 0 to 3.8 atmospheres; at 17.93 per cent the surface force was 11.4; at 10.06 per cent, 22.4; and at 13.16 per cent, about the hygroscopic coefficient, the surface force increased to 72 atmospheres. Thomas[49] measured the aqueous vapor-pressure lowerings of several soils at different moisture contents. One of the loam soils used by him had a moisture equivalent of 23.3 per cent. By growing beans, Thomas found the plants wilted in this soil at 9.3 per cent. This gives a ratio of the moisture equivalent to the moisture content when the beans wilted of 2.5, which is much higher than the ratio of 1.84 given by Briggs and Shantz.[12] The calculated wilting coefficient for this soil, using the ratio of 1.84, is 12.7 per cent. Thomas[49] reports a vapor pressure depression of 0.30 mm. at a moisture content of 9.5 per cent for this soil, which in a solution at 25° means an osmotic pressure of about 17 atmospheres. The vapor pressure depression at 12.7 per cent calculated from Thomas' formula would be 0.14 mm., which means an osmotic pressure of about eight atmospheres, which is about twice that found by Shull. Thomas' data indicates that above the calculated wilting coefficient the vapor pressure changes are very slight for large variations in moisture con-

tent but that the vapor pressure begins to fall rapidly with lower moisture contents.

It has been shown by a number of investigators that plants do not absorb solutes in the same proportion in which they exist in the soil solution. It also appears that the amount of solutes absorbed bears no direct relation to the amount of water absorbed, and that increasing transpiration does not accelerate the entrances of solutes. Hoagland[33] has recently pointed out that plants can make equally good growth in a very great variety of culture solutions and that there is no evidence that plants thrive only in solutions with certain specific ratios between various elements. Solutes and solvent may move into the plant independently and the movement of water within the plant may be independent of the movement of solutes, as Curtis[24] seems to show, and since there is a wide range of soil solutions in which plants develop normally, there seems to be no physiological reason why the environmental soil conditions necessary for good growth should not be met within a range of moisture content from the maximum field, or capillary capacity, to about the wilting coefficient. Furthermore, the fact that the best evidence seems to show that the force with which water is held by the soil does not increase rapidly until the moisture content is reduced below the wilting coefficient indicates that optimum moisture conditions extend from the field capacity to about the wilting coefficient.

There was close agreement between the wilting of these prune trees without recovery until water was added to the soil and the calculated wilting coefficient of the loam soils used in these experiments. Inasmuch as these observations were made at Mountain View and Davis throughout several seasons and at different times during each season there were differences in atmospheric evaporation power, and yet the wilting coefficient of these loam soils calculated from the moisture equivalent seemed to be a measure of a moisture condition at which the prune trees wilted and did not recover until water was added to the soil.

The moisture content of the loam soils in the tanks was reduced much below the calculated wilting coefficient by the prune trees but in no case was the water supply reduced to the calculated hygroscopic coefficient. The trees were able to use only about half of the water in the soil between the wilting coefficient and the hygroscopic coefficient. Substantially the same results were obtained in the Santa Clara Valley prune and the Muir peach orchard irrigation studies. However, the soil-moisture supply was reduced to the hygroscopic coefficient in a few cases by these mature trees.

Vetch plants grown in tanks were allowed to reduce the moisture content of the soil. When no further loss of moisture could be detected by the means at hand for weighing the tanks, samples taken indicated that approximately half of the moisture between the wilting coefficient and the hygroscopic coefficient had been used. Although the wilting coefficient is not the lower limit of available water, it probably is a better basis for comparing moisture conditions of soils than the hygroscopic coefficient which probably represents the extreme lower limit of available moisture but which, according to Puri,[43] can not be satisfactorily determined.

The different amounts of water available in the soil apparently had no noticeable effect on the condition, in the fall, of the prune trees grown in tanks. Defoliation caused by wilting could be brought about by withholding water until the soil-moisture content was reduced to the wilting coefficient, but no differences could be noted in the time the leaves normally dropped when trees on soil with high moisture content were compared with those on soils with lower amounts of available water, but in which the soil-moisture content was not reduced below the wilting coefficient. The fact that the time of beginning of the coloring of the leaves in the fall, and probably of the beginning of dormancy or maturity of the plant tissues, was not affected by variation in amounts of available soil moisture within a wide range, should be expected from the results reported in the first part of this section, wherein it is shown that the use of water and the growth of these young prune trees were not affected by variations in amounts of available water during the growing season.

While it must be remembered that the results of the studies under controlled conditions were obtained with young prune trees in containers, and that they are applicable only under the conditions of these experiments, they do suggest that many of the current views regarding the soil-moisture relations of other plants may also be questioned.

SECTION IV

THE LOSSES OF MOISTURE BY EVAPORATION DIRECTLY FROM THE SURFACE OF THE SOIL AND THE MOVEMENT OF MOISTURE IN THE SOIL

Early in the course of the studies described in the preceding sections, it was noted that the amount of water transpired by growing plants was tremendously greater than that evaporated directly from the surface of the soil. These results are based not only on comparisons of the losses of water through trees growing in tanks with losses of water from uncropped tanks, but also on the results of sampling the soil for moisture determinations in fallow and cropped areas in the field. Therefore, if it is established that the great majority of water is taken from the soil by plant transpiration, the results of sampling the soil in orchards, if such sampling be representative, might indicate the presence or absence of roots, which in turn would indicate the depth of soil necessary to be wetted at each irrigation. For this reason, as well as for the direct bearing the results of such studies have on orchard practices, and the importance of information concerning the movement of moisture in soils, the studies reported in this section were undertaken. Furthermore, the absence of rains during the summer months in California makes the conditions unusually favorable for such studies and affords an opportunity not generally met with in other sections.

The conclusion that losses of moisture are occasioned by the upward movement of moisture from the lower moist layers of soil to the surface and dissipated into the air is based upon the familiar teaching that moisture is capable of moving in the soil in all directions through capillary forces. It is reasoned that moisture exists in the form of films around the soil particles and in wedge-shaped masses of water between the soil particles at the points of contact with each other, being held partly by the attraction of the non-water particles

for the water, and partly by the molecular attraction of the liquid itself. Adjoining particles with different thicknesses of moisture films surrounding them hold different amounts of water in the adjacent capillary spaces.

The amount of this capillary or imbibitional water in the soil is determined by a number of conditions. The most important of these is the number of soil particles, the soil texture, and the arrangement of the particles, or the structure of the soil. The depth of the soil to the level of standing water also is an important factor in determining the amount of water held in the soil. Other conditions, which exert much less influence, are the temperature and the kind and quantity of material dissolved in the water.

Briggs[11] points out that the moisture surface around the particles containing less amounts of water than adjacent particles will have greater curvature and consequently greater pressure outward. Moisture will move through the connecting film from the greater to the lesser mass until the pressure becomes the same, a process which takes place when the capillary spaces contain equal amounts of water. This movement can extend to any number of capillary spaces through any number of films. A decrease in the amount of water in the capillary spaces in the upper layers of soil would then be expected to result in water being drawn to that point.

However, Buckingham[17] pointed out many years ago that when a soil is very wet, the water in the capillary spaces is continuous and the wedge-shaped masses merge into one another at their edges. A current of water could flow through the soil without having to flow through the films. As the water content of the soil is reduced, the capillary masses or drops cease to be continuous and communicate with one another only through the film water. Then the resistance of these thin films to the flow of water is much greater than the resistance in the parts of the path which lie in the capillary water, and it is reasonable to assume that the movement is materially retarded.

The commonly accepted idea of the part that cultivation plays in preventing the upward rise and consequent loss of moisture into the atmosphere is based upon the theory that by loosening the surface, rapid drying takes place and the soil particles are removed from close contact with each other. The first condition results in a decrease in evaporation, since the dry soil is assumed to act as a blanket; the second, reducing the points of contact, lessens the capillary pulling power.

The movement of moisture in soils contained in glass or metal cylinders and columns of soil in tanks or metal lined flumes, when the lower ends are in contact with free water, has received much study. The movement of moisture from moist soils to soils containing lesser amounts of moisture, the field condition more frequently met with in California, has received less study. The work of Harris and Turpin,[30] Alway and McDole,[3] Willard and Humbert,[54] and McLaughlin,[40] is typical of many of the experiments dealing with moisture movement under controlled conditions in soils where a water table is not present. It might be mentioned in passing that the results obtained by these investigators all indicate, although some of them did not come to this conclusion, that the movement of moisture, especially in an upward direction, from moist soils to drier soils is slight in amount and extent, when the source of the moisture supply is not saturated soil in contact with a free water surface.

Numerous observations of moisture conditions in the field by soil sampling have been reported and, in many cases, the upward movement of moisture by capillary action, and consequent large losses by evaporation have been held to be extremely important. Widtsoe[53] states that there is, soon after each irrigation, a steady upward movement of water causing a general drying out of the soil to great depths. Cameron[20] maintained that the larger part of the water from rains in humid areas returns to the surface through distances of many feet. Hall[29] holds that in some soils the capillary rise of water might be as much as 200 feet.

King,[37] in many of the reports of the Wisconsin Agricultural Experiment Station and in his several text books, laid stress upon the losses of moisture through evaporation from the soil surface by upward capillary movement. Hilgard and Loughridge,[31] in the reports of the California Agricultural Experiment Station, held the same opinion. In the reports for 1897–1898 of this station, cultivation to conserve soil moisture under semi-arid and arid conditions and the need for deeper cultivation under these conditions are strongly recommended. Fortier and Beckett[26] studied the losses of moisture from irrigated soils contained in large tanks and state that soil mulches are effective in lessening these losses.

On the other hand, Alway[2] concludes that but comparatively little water which has once passed below the first foot is lost by evaporation, and again the same investigator[1] concludes that the loss of moisture from the subsoil of dry lands under crop seems to take place almost entirely through transpiration. From field studies extending

over seven years, Burr[16] reached the conclusion that in order to obtain water the roots of plants must extend themselves into the soil where available water is present, rather than depend upon the water being brought to them by capillarity, since there seemed to be little upward movement of subsoil water in the absence of a water table.

Loughridge,[39] in reporting observations of moisture movement in citrus groves, lays stress on the loss of moisture by evaporation and upon the value of cultivation, but shows that there is relatively little lateral movement of moisture from irrigation furrows when the downward movement is not obstructed. Moreover, his data indicate that no loss of moisture by either upward or downward movement occurred below four feet in a citrus grove through the period of one month. He states, "It is extremely doubtful that water at a depth of more than five feet below the root systems will be of any benefit to the trees in times when needed, for the capillary rise is extremely slow." Briggs, Jensen and McLane[15] found that available moisture below the third foot did not prevent orange trees from wilting when the moisture content of the upper soil which contained the roots of the trees had been exhausted below the wilting coefficient.

In studying the root systems of corn and sorghum in Kansas, Miller[41] found that there was little depletion of soil moisture below the depth to which the roots penetrated. Baker[4] reports that the loss of moisture by direct evaporation from the surface of the soil is very small after the water has become distributed. It is reported by Young[55] that the loss of moisture is largely by transpiration through plants. He also concludes that the soil mulch is not more effective than an unmulched soil in retarding evaporation. Call and Sewell[19] conclude that cultivation is not effective in preventing evaporation.

Rotmistrov[44] makes the following statement after a number of years' observation, "As regards the circulation (of moisture) in an upward direction, there exists a wrong impression, which our literature has almost made a household word. It is maintained that water can rise to the surface from the deep layers by capillary action. I shall not name the authors who maintain this theory—they are too numerous—but I do not know of a single author who could prove this proposition," and again "that water percolating beyond a depth of 40 to 50 centimeters does not return to the surface except by way of roots."

Sewell[45] reviewed much of the literature dealing with tillage, and since tillage is supposed to affect soil moisture, his conclusion, that cultivation may be necessary only to kill weeds and to keep the soil

in a receptive condition to absorb rainfall, may be of interest in this connection. Furthermore, Chilcott and Cole,[21] after an extensive investigation in the Great Plains, conclude, "The quite general popular belief in the efficiency of deep tillage as a means of overcoming drought, or of increasing yields, has little foundation of fact, but is based on misconceptions and lack of knowledge of the form and extent of the root systems of plants, and of the behavior and movement of water in the soil."

Fig. 29. Soil tanks at Mountain View used to determine the losses of moisture by evaporation directly from the surface of the soil. Later the tanks were further protected from temperature changes by pieces of board sheathings cut to fit around the tanks.

Losses of Moisture by Evaporation Directly from the Surface of Bare Soils in Tanks

Tanks similar to those in which the trees were planted were used during the summer of 1921 at Mountain View, in tests to determine the loss of water from bare soil by evaporation directly from the surface. These tanks were of two sizes: the smaller tanks with an exposed surface of 3.01 square feet held about 1000 pounds of soil; the larger tanks with an exposed surface of 3.69 square feet held about 1400 pounds of soil. The tanks, all of which were 48 inches deep, were packed with a Yolo clay loam soil, and the packing was such that it had the same volume weight as that of the field soil. The tanks were allowed to stand for six months before the tests were begun. The weight of water-free soil in each tank was known.

On August 17, 1921, sufficient water was applied to all of the tanks except Nos. 23, 24, and 26, to equal a calculated average percentage of moisture in the soil of 20 per cent. It was thought undesirable to disturb the soil in the tanks; therefore, no samples were taken immediately after the water was applied. As a consequence, the distribution of water in the soil in the tanks at this time is not known. However, the amount of water applied should have been sufficient to wet all of the soil to within six inches of the bottom. More than enough water was applied to tanks Nos. 23, 24, and 26 to fill the soil to the maximum field capacity. The excess water was drained off during the first five days after the water was applied. These tanks, therefore, were wet throughout the entire depth of soil.

The soils in nine of these tanks, Nos. 23, 25, 27, 29, 31, 33, 36, 38, and 41, were not disturbed after the water was applied to the surface, except to pull carefully any weeds which started to grow. The soils were cultivated to a depth of six inches in the tanks numbered 24, 26, 30, and 39, as soon as possible after the water was applied. The soils in the tanks numbered 28, 32, 35, and 42 were cultivated to a depth of eight inches, and the soils in the tanks numbered 34, 37, and 40 were cultivated to a depth of ten inches. The soils were cultivated every week until the end of the test by means of a small five-pronged garden fork, which was thrust into the soil the desired depth, thus loosening and stirring it. This was followed by further cultivation with a small pointed hoe shaped somewhat like the blade on a shovel cultivator. This hoe was drawn backwards and forwards through the soil, thoroughly stirring it. The tanks were weighed at frequent intervals during the course of the test, which ran from August 17, 1921, to November 4, 1921. The weighings were made with the

portable derrick and suspension scales previously described. The arrangement of the tanks in the trench and the condition of the surface of the soil in some of the tanks is illustrated in figure 29, the photograph for which was taken on August 24, 1921, seven days after the water was applied. Later, the tanks were further protected from undue temperature changes by pieces of board sheathings cut to fit around the tanks.

The accumulated loss from the beginning of the tests, August 17, 1921, until November 4, 1921, at different times as determined by weighing with the portable derrick and suspension scales is given in table 29. Since the tanks differed in size, the loss is calculated as pounds to the square foot of surface exposed to evaporation. One pound of water to a square foot is equivalent to a depth of 0.19 inches, or about $\frac{3}{16}$ inches, of water. The data in table 29 are shown graphically in figure 30.

It will be noted that in every case the loss which occurred within the first week after the water was applied was approximately 50 per cent of the total loss which occurred within 80 days. The relatively rapid loss immediately after the application of water to the soils in the tanks is shown by the results obtained from observations made in 1922 on tanks Nos. 26 and 27, containing Yolo clay loam, with an area exposed to evaporation of 3.01 square feet. These tanks were irrigated on May 14, 1922, with sufficient water to saturate the bottom six inches of soil in the tanks. The soil in tank No. 26 was found to contain an average moisture content of 23.9 per cent, and the soil in tank No. 27 contained 24.0 per cent moisture when samples were taken on October 25, 1922, 164 days after the water was applied. Tank No. 26 lost 14 pounds, and tank No. 27 lost 13 pounds during the week following irrigation. The accumulated loss at the end of the first month from tank No. 26 was 20 pounds, and from tank No. 27, 19 pounds. The total loss for the entire 164 days, or from May 14 to October 26, 1922, from tank No. 26 was 35 pounds and from tank No. 27, 32 pounds. The surface of the bare soil in these tanks were undisturbed except to pull weeds which started to grow.

These rapid losses by evaporation from the soil surface, which immediately followed irrigation and which constituted about one-half of the total loss from this cause within a period of about three months, occurred before the surface of the soil was in condition to be properly cultivated. It will be seen, then, that the supposed efficiency of cultivation in controlling these losses is doubtful in any event, since so large a portion of the total loss occurs before the surface of the soil is dry enough to be properly cultivated.

TABLE 29

LOSSES OF MOISTURE IN POUNDS TO THE SQUARE FOOT* BY EVAPORATION DIRECTLY FROM THE SURFACE OF BARE SOILS IN TANKS CONTAINING YOLO CLAY LOAM AT MOUNTAIN VIEW. WATER WAS APPLIED TO THE SOIL ON AUGUST 17, 1921. THE TOTAL LOSSES UP TO EACH DATE ARE GIVEN

Tank	Treatment of soil	Area of soil in tank exposed to evaporation, sq.ft.	August			September					Oct.	November	
			25	27	29	2	10	16	24	30	8	1	4
23		3.69	4.0	4.9	5.4	6.2	7.3	8.4	8.4	9.2	9.2	10.3	10.3
25		3.01	4.0	4.6	4.6	5.6	6.0	6.6	7.0	7.6	7.3	8.0	8.3
27		3.01	4.6	4.6	5.3	6.0	6.6	7.0	7.3	8.1	7.6	8.6	8.6
29	Undisturbed	3.69	4.9	5.2	5.9	6.2	7.3	8.1	8.1	9.0	9.2	10.0	10.0
31	except to	3.01	5.6	6.0	6.3	7.0	7.6	8.0	9.0	9.3	9.0	10.0	10.0
33	pull weeds	3.01	5.3	6.0	6.3	7.3	7.6	8.0	8.6	9.0	9.0	9.6	9.6
36		3.01	3.6	4.0	4.3	5.0	6.0	6.3	7.3	7.3	8.0	8.3	8.6
38		3.01	4.3	5.0	5.0	6.0	6.6	7.3	7.6	8.0	8.3	10.0	9.6
41		3.69	4.6	4.6	5.4	5.7	6.5	6.8	7.0	8.4	8.4	9.2	9.2
	Average		4.6 ±0.15	5.0 ±0.15	5.4 ±0.15	6.1 ±0.13	6.8 ±0.15	7.4 ±0.2	7.8 ±0.16	8.4 ±0.16	8.4 ±0.13	9.3 ±0.19	9.4 ±0.17
24	Cultivated	3.69	4.3	4.8	6.0	6.2	7.6	7.8	8.2	9.0	8.6	10.0	10.8
26	weekly to	3.01	5.0	5.6	5.6	6.4	7.6	7.6	8.6	9.4	9.4	10.3	10.3
30	depth of	3.69	4.1	4.0	4.8	5.4	6.8	6.8	7.0	7.6	7.6	8.9	9.2
39	6 inches	3.01	4.6	5.0	5.0	5.4	6.4	6.6	7.0	8.0	8.0	8.2	9.0
	Average		4.5 ±0.13	5.0 ±0.22	5.4 ±0.2	5.8 ±0.18	7.0 ±0.22	7.2 ±0.2	7.8 ±0.28	8.4 ±0.19	8.4 ±0.24	9.4 ±0.31	9.8 ±0.29
28	Cultivated	3.01	5.3	5.4	6.0	6.4	7.0	7.6	8.0	8.4	8.4	9.6	10.0
32	weekly to	3.01	5.0	5.6	5.6	6.6	7.0	7.4	8.0	8.0	8.4	9.6	10.0
35	depth of	3.69	3.8	4.0	4.4	4.8	6.0	6.6	7.0	7.0	7.0	8.4	8.4
42	8 inches	3.69	5.2	5.4	5.7	6.0	6.8	7.4	7.6	8.2	8.6	9.5	9.5
	Average		4.8 ±0.23	5.0 ±0.24	5.4 ±0.25	6.0 ±0.15	6.6 ±0.16	7.2 ±0.17	7.6 ±0.15	7.8 ±0.19	8.0 ±0.24	9.3 ±0.20	9.5 ±0.25
34	Cultivated	3.01	4.3	5.0	5.1	5.3	6.0	6.6	6.6	7.0	7.3	8.1	8.7
37	weekly to	3.01	4.0	5.0	5.0	5.3	6.4	6.6	7.0	7.4	7.4	8.6	8.6
40	depth of 10 inches	3.01	4.6	4.6	5.0	5.2	6.0	6.7	7.4	7.4	7.3	8.6	8.7
	Average		4.3 ±0.11	4.9 ±0.08	5.0 ±0.03	5.4 ±0.03	6.0 ±0.05	6.6 ±0.03	7.0 ±0.16	7.3 ±0.09	7.3 ±0.03	8.4 ±0.10	8.7 ±0.03

* One pound of water to a square foot is equivalent to 0.19 inches of water.

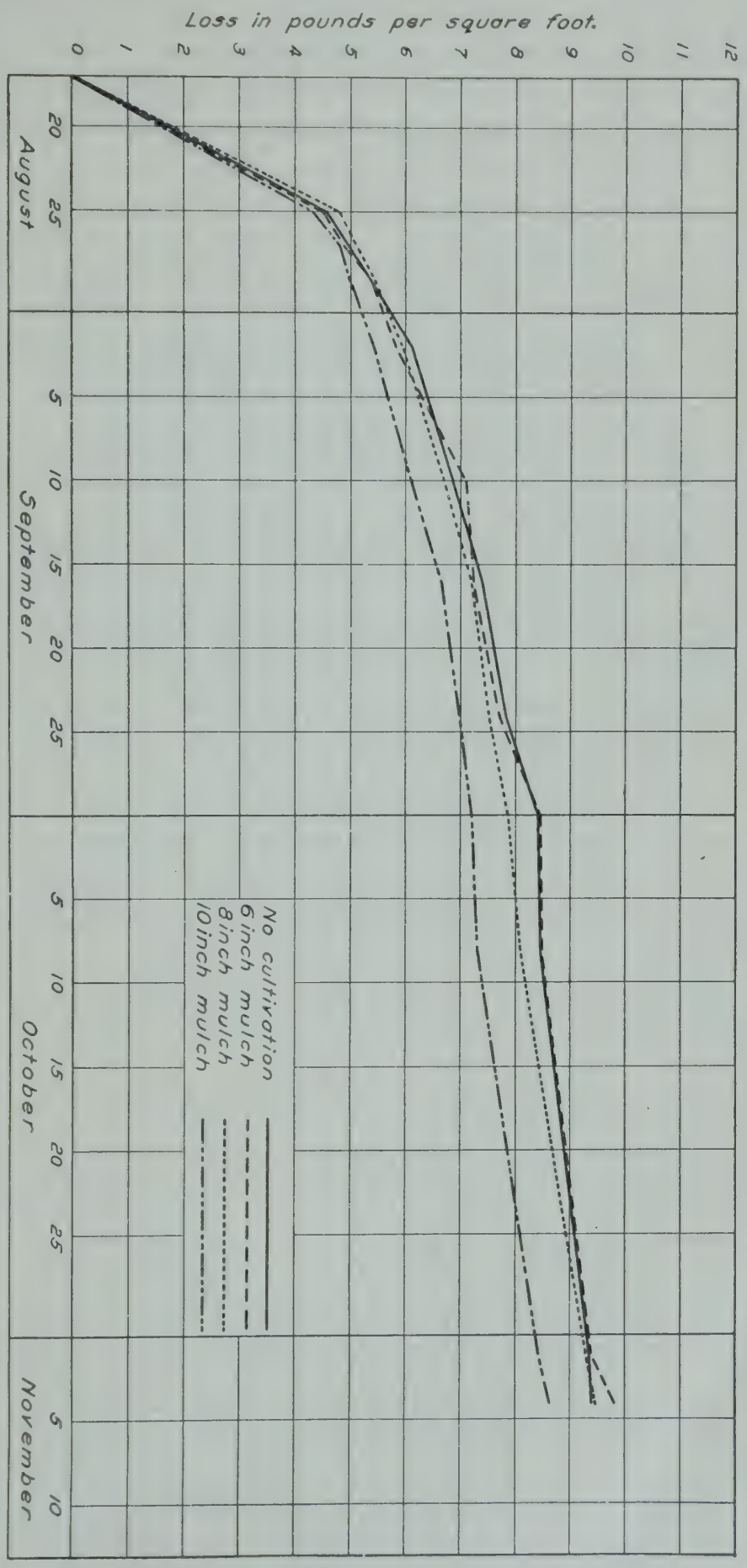


Fig. 30. Loss of water by evaporation directly from the surfaces of bare soils in tanks at Mountain View, 1921.

Several of the tanks were kept under observation for long periods of time. One of these, tank No. 25, which may serve to illustrate the extremely slow rate of loss of moisture after the first few weeks, had an area exposed to evaporation of 3.01 square feet. This tank was irrigated on August 17, 1921, and the bare surface of the soil was undisturbed except to pull the weeds which started to grow. The tank was covered during rains to prevent the addition of water to the soil in the tank after the initial irrigation. The losses from this tank for the period of August, 1921, to November, 1922, are graphically shown in figure 34. The total accumulated loss of moisture by evaporation at the end of different periods of time is given in table 31. On November 2, 1925, the total loss was 57 pounds, which is 18.9 pounds to the square foot of surface exposed to evaporation, and is equivalent to a depth of about 3⅜ inches of water in a period of over four years. The data (see table 31) when compared with those presented in Sections I and II show that amounts of water equivalent in depth to this were used very rapidly by mature prune and peach trees. This tank was at Mountain View up to February, 1923, and was then moved to Davis.

When the average losses of moisture by evaporation from the uncultivated soil and the cultivated soils are compared, and the probable errors of the mean values are considered, it will be seen that there are no significant differences in the results obtained. Furthermore, the average loss of water by evaporatsion directly from the surface of the bare uncultivated tanks in 80 days was 9.4 ± 0.17 and the average loss from the eleven cultivated tanks was also 9.4 ± 0.16. It is apparent, then, that cultivation seems to have little influence in controlling the amount of water evaporated from the bare surface of the soil in the tanks.

Other comparisons between the losses of moisture from cultivated and uncultivated soils were made at Davis. Some of the tanks used in this way at Davis are shown in figure 21. The results have been the same as those obtained at Mountain View, in that apparently there are no real differences between the losses from the cultivated and uncultivated soils. During 1923 and 1924, a soil much lighter in texture than the soil previously used either at Davis or at Mountain View was tried. A sandy loam, with a moisture equivalent of 15.7 per cent was packed in the tanks. The losses of moisture by evaporation from the surface of bare soils were determined for a period of 11 months. The average loss of moisture by evaporation during this time was 51 pounds to a tank, or 13.8 pounds to the square foot of area exposed to evaporation. This total loss is greater

than the loss from finer textured soils, such as the Yolo clay loam or the Yolo loam. The evaporation losses observed from field plots were also greater on the sandy soil.

TABLE 30

PERCENTAGE OF MOISTURE IN SOIL IN TANKS AT MOUNTAIN VIEW. SAMPLES TAKEN NOVEMBER 4, 1921. TANKS IRRIGATED AUGUST 17, 1921

Tank	Treatment of soil surface	Depth of soil samples (inches)					Depth of soil samples (inches)			Totals
		0–4	4–8	8–12	12–16	16–20	0–12	12–24	24–26	0–36
23		9.8	16.1	18.4	20.4	22.5	13.4	20.6	24.0	19.2
25		11.3	15.9	17.8	17.3	18.9	13.2	18.4	19.7	16.9
27		10.5	14.9	17.0	18.9	20.0	18.4	18.7	17.6	17.9
29	Undisturbed	9.7	16.0	18.0	19.7	17.7	16.2	18.6	18.6	17.8
31	except to pull	8.8	15.5	16.9	18.5	18.5	13.7	19.4	18.1	17.1
33	weeds	8.2	15.5	16.5	18.8	18.6	13.3	18.7	18.1	16.7
36		8.7	17.2	18.1	19.4	18.4	13.0	17.4	17.1	16.0
38		9.3	13.1	18.3	18.3	17.0	11.3	17.7	18.4	15.5
41		8.3	13.3	18.2	20.2	19.1	12.2	19.4	17.0	15.6
	Average.....	9.4	15.3	17.7	19.2	19.0	13.9	18.8	18.7	17.0
		±0.25	±0.30	±0.18	±0.24	±0.32	±0.46	±0.21	±0.41	±0.28
24	Cultivated	10.0	19.3	18.6	20.0	20.2	15.7	19.0	22.6	19.2
26	weekly to	10.7	18.9	20.3	21.3	21.7	14.8	20.5	24.0	19.5
30	depth of	10.0	21.9	19.0	18.7	19.9	15.6	21.6	17.0	17.8
39	6 inches	8.0	15.5	17.7	15.9	20.7	15.3	.....	16.6	.....
	Average.....	9.7	18.9	18.9	19.0	20.6	15.3	20.4	20.0	18.8
		±0.40	±0.83	±0.37	±0.82	±0.28	±0.15	±0.54	±1.59	±0.46
28	Cultivated	10.4	12.5	18.9	11.7	20.0	14.6	20.2	18.0	17.8
32	weekly to	8.6	16.7	18.7	19.3	19.5	14.0	18.0	16.2	16.2
35	depth of	7.0	16.6	17.2	19.4	19.3	11.0	19.1	18.8	16.7
42	8 inches	6.7	14.9	18.1	18.4	20.4	12.6	20.2	15.5	16.5
	Average.....	8.9	15.2	18.2	17.2	19.8	13.0	19.4	17.1	16.8
		±0.72	±0.72	±0.28	±1.34	±0.20	±0.61	±0.40	±0.61	±0.24
34	Cultivated	8.8	16.0	18.2	20.0	20.1	12.9	19.0	19.5	17.7
37	weekly to	7.0	14.2	17.4	16.6	17.1	12.7	15.4	17.6	15.3
40	depth of	6.7	14.1	15.1	17.6	15.1	13.4	17.9	17.1	16.4
	10 inches									
	Average.....	7.5	14.8	16.9	18.1	17.4	13.0	17.4	18.1	16.5
		±0.52	±0.50	±0.72	±0.78	±1.06	±0.16	±0.82	±0.57	±0.50

Distribution and Total Amount of Moisture in Soil Following Exposure to Evaporation

Samples to determine the amount and distribution of moisture in the soil were taken on November 4, 1921, 80 days after the water was applied, in all of the tanks for which data are reported in table 29. Samples of the soil in 4-inch layers to a depth of 20 inches were taken with a special large soil tube and samples in foot depths to 3 feet were also taken. The results of these moisture determinations are given in table 30.

Apparently there is little difference in distribution of moisture in the soil in the cultivated and uncultivated tanks. The greater amount of loss of moisture seemed to be confined largely to the upper 4 inches of soil. It will be noted that below 8 inches there were very uniform percentages of moisture, even 80 days after the water was applied.

TABLE 31

Distribution of Moisture in the Bare Uncultivated Soil in Tank No. 25. Water was Applied to the Soil on August 17, 1921*

Depth of soil samples, inches	Moisture equivalent	Dates samples were taken					
		Nov. 4, 1921	Apr. 11, 1922	June 20, 1922	Dec. 15, 1922	Aug. 8, 1924	Nov. 2, 1925
0 to 4	21.5	11.3	7.6	.....	6.8	5.3	4.3
4 to 8	22.6	15.9	13.8	.....	11.5	5.7	6.2
8 to 12	22.3	17.8	15.4	.....	13.4	10.2	9.8
12 to 16	23.1	17.3	16.6	.....	14.9	11.1	11.4
16 to 20	22.5	18.9	17.3	.....	16.3	13.0	11.8
Average	22.4	16.3	14.2	.....	12.6	9.2	8.7
0 to 12	22.0	13.2	12.0	10.8	9.8	6.3	6.1
12 to 24	21.9	18.4	17.7	18.0	15.6	10.6	11.6
24 to 36	22.7	19.7	19.6	17.0	18.0	15.5	15.6
36 to 42	22.3	.....	.....	.....	19.0	16.0	16.0
Average	22.2	17.1	16.4	15.3	15.5	12.1	12.3
Pounds lost since water was applied on Aug. 17, 1921		25	35	35	37	51	57

* The average moisture content calculated from the weight of dry soil in the tank was 20 per cent on August 17, 1921. However, the soil was not wet to full depth. The tank was covered during rains and no water was added after the application on August 17, 1921.

Tank No. 25, from which the evaporation losses have been measured for a long period of time, was sampled several times during the three years of these observations. The results of these moisture determinations are given in table 31. There was a gradual loss at an extremely slow rate throughout the entire depth of soil. This result was also noted in other tanks observed for long periods; however, it is remarkable that even after four years' exposure to evaporation, the average moisture content of all of the soil in the tank was not reduced below the wilting coefficient. Whether the losses of moisture from the lower depths of soil were due to upward capillary movement, or to the gradual drying effect of the movement of air through the soil mass is not known. This point is discussed further in subsequent pages.

The presence of relatively large amounts of water in the soil in these tanks, even after long exposure to evaporation was demonstrated in a rather striking way. Sixteen of the 20 tanks used in the evaporation experiment during the summer of 1921 were planted to vetch and barley after having been exposed to evaporation for 80 days, namely, on November 4, 1921. Since, as indicated in table 30, the first 4 inches of soil were too dry to germinate the seed, from 3 to 4 pounds of water (less than one-half gallon) were added to the tanks after planting. Owing to a dry hot wind which followed the planting and dried out the soil before the seeds germinated, a few of the tanks received a subsequent application of a like amount of water. After the plants were up, they were thinned to a definite number to a tank. The number selected gave a stand comparable to that usual with cover crops in orchards. It was possible to mature satisfactory cover crops in these tanks. The loss of moisture by evaporation during the previous 80 days, which is longer than the usual time between irrigations, had not been sufficient to prohibit the growth of cover crops. The tanks were covered during rains by means of a canvas drawn over a frame. The tanks containing the vetch and barley plants are shown in figure 31, as they were on March 10, 1922. This figure should be compared with figure 29, in which the same tanks are shown.

Both the vetch and barley made the same growth in all of the tanks, and no difference could be distinguished between the growth made by the plants in the tanks which had been cultivated and those which had not been disturbed during the 80 days preceding the planting of the seeds. Apparently, cultivation had not affected the amount of water available for growth or its distribution in the soil.

The growth made by the vetch in tank No. 36 is shown in figure 32. The soil in tank No. 36 was undisturbed for 80 days after the irrigation on August 17, 1921, and was in the condition shown in tank No. 35, figure 33. The growth made by the vetch plants in tank No. 30 is shown in figure 33. The soil in tank No. 30 was cultivated during the 80-day period and was in the condition shown in tank No. 29, figure 33. Tank No. 30 was larger than tank No. 36 and contained more plants, otherwise the growth and vigor of the plants was the same. The photographs were taken on May 6, 1922.

The Yolo clay loam in these tanks had a relatively high moisture-holding capacity and the plants were grown for the greater part of the time when the transpiration losses were low. It is, of course, obvious that such an experiment could not be carried on with a sandy soil, or one with a low moisture-holding capacity, or during a time of the year when the atmospheric evaporating power was high. However, it does clearly show that there was a relatively large quantity of water left in the soil after a long period of exposure to evaporation and that there were no differences in available moisture in the soils which had been cultivated and those which had not been cultivated during this period.

The loss of water from four typical tanks used in these experiments is graphically illustrated in figure 34. The results obtained from tanks Nos. 30 and 36, which have just been described, are also shown in this figure. The increased loss after the vetch was planted in these tanks on November 4, 1921, is clearly shown. The total loss from tank No. 30, from August 17, 1921, to June 1, 1922, when the vetch was matured was 148 pounds. The loss from August 17, 1921, to June 30, 1922, from tank No. 36, which contained a fewer number of plants, was 110 pounds. The loss from August 17, 1921, to June 20, 1922, from tank No. 25, the bare uncultivated tank, was only 35 pounds. The loss was 39 pounds from tank No. 32, the bare cultivated tank during this time.

After the crop of vetch was removed from tank 36, on June 20, 1922, it was irrigated and vetch was again planted. On September 11, 1922, this crop matured. During this time 295 pounds of water were used. The vetch was irrigated several times. The loss from the bare uncultivated tank No. 27, at this time of the year, is shown for comparison in figure 34. The loss from tank No. 36 after the crop was removed on September 11, 1922, and after it was again irrigated, is also shown.

Fig. 31. Barley and vetch plants growing in tanks irrigated 80 days before the seeds were planted. Rain was prevented from wetting the tanks by means of the wooden frame and a canvas cover. Photographed March 10, 1922.

Fig. 32. Vetch plants growing in tank No. 36 on soil which had been exposed to evaporation for a period of 80 days after irrigation and before the seeds were planted. The soil in this tank was uncultivated during this period, being in the same condition as that in tank No. 29, shown on the left in figure 33. Photographed May 6, 1922.

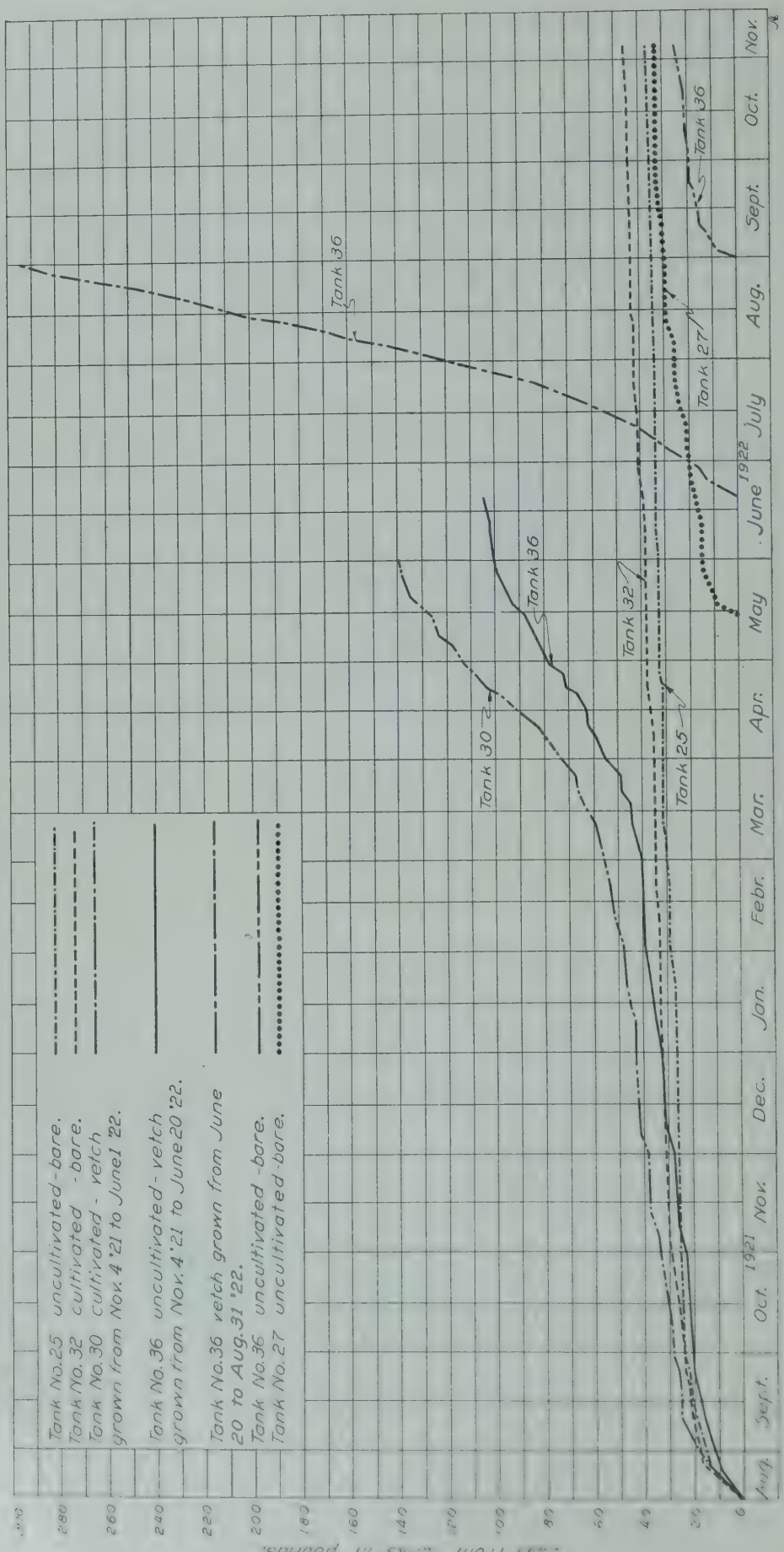


Fig. 34. Loss of water from bare soils compared to the loss through plants.

Several tanks, among which was tank No. 27, for which the evaporation loss is shown in figure 34, were irrigated on May 13, 1922, and the surface soil was undisturbed until October 26, 1922. During this time tank No. 27 lost 32 pounds through evaporation. On this date the soil to a depth of one foot in these several tanks was removed and thoroughly mixed on a piece of canvas and replaced in the tanks. Vetch seedlings, which had been grown in flats, were planted in the tanks. No water was applied to the soil and the tanks were protected from the rains. The vetch plants matured, making a growth comparable to that of similar plants in the orchards even though the uncultivated soils had been exposed to evaporation from May 13, 1922, to October 26, 1922, a period of 167 days.

Fig. 33. Vetch plants growing in tank No. 30 on soil which had been exposed to evaporation for a period of 80 days after irrigation and before the seeds were planted. The soil in tank No. 30 was cultivated during this period and was in the same condition as that in tank No. 35, shown on the left in figure 32. Photographed May 6, 1922.

Losses of Water from Bare Soils Compared to Losses Through Plant Transpiration

The data graphically illustrated in figure 34, details concerning which have been described, and the instances mentioned above, show that the losses by evaporation directly from the surface of the soil are a small portion of the total losses which occur from irrigated soils on which plants are growing during the summer.

The records obtained from the tanks in which trees were growing also afford an opportunity to compare the losses by evaporation

directly from the soil surface and those through transpiration. While it can not be assumed that such comparisons are strictly quantitative they do serve to show that the losses by evaporation from the soil are extremely small when compared to the amount of water transpired by the trees. In 1921, a three-year-old prune tree in a tank used 585 pounds of water between May 21 and November 21, a period of six months, while a similar tank containing the same kind of soil with a high moisture content but uncropped, lost only 28 pounds of water within the same time, an amount no greater than this small tree used in one instance within three days. This same tree, from March 1, 1922, to November 4, 1922, used 1250 pounds of water, while the uncropped tank, which was irrigated February 11 with 23 pounds of water, again lost only 28 pounds of water between this date and November 4, 1922. Tank No. 2, the bare uncultivated tank just mentioned, contained Yolo loam, which had been packed in the tank in 1912, and had not been disturbed since that time.

A further illustration of the demand made upon the soil-moisture supply by growing plants is shown in the record of the use of water by morning glory (*Convolvulus arvensis*) plants grown in a tank. This tank was 23½ inches in diameter and 48 inches in depth, and was packed with Yolo clay loam at Mountain View, in February, 1921. It was one of the series used to determine the losses of moisture by evaporation directly from the surface of the soil in which the soil was cultivated to a depth of six inches following the application of water on August 17, 1921. The loss of water by evaporation from this tank for the first 80 days after irrigation, given in table 29, was 31 pounds, or 10.3 pounds to a square foot. The total loss of moisture by evaporation from August 17, 1921, to May 13, 1922, the date on which water was again added to the soil, was only 41 pounds or 13.6 pounds to a square foot. The tank was irrigated again on May 13, 1922, but the surface soil was undisturbed until October 26, 1922. Between May 13 and October 26, 1922, only 35 pounds of water were lost by evaporation. Vetch plants were set on October 26, 1922, and grew to maturity without additional water being applied in this tank as they did in tank No. 27, which has been described. The vetch was taken out of the tank in June, 1923, and the soil was undisturbed throughout the summer.

Three pieces of morning glory (*Convolvulus arvensis*) roots were planted in the tank on October 11, 1923, and the soil was thoroughly irrigated. Early in the spring of 1924, the roots began to grow, and by March 28, 1924, leaves had begun to appear. The soil was irri-

gated 10 times up to August 19, 1924. Between March 28, 1924, and August 19, 1924, a period of 144 days, the tank lost 704 pounds of water. The appearance of the plants on August 19, 1924, is shown in figure 35. It will be seen that the surface of the soil was well shaded by the plants; there was probably little loss of water by direct evaporation from the soil surface. The record of the loss of water from the young plants in this tank, for a period of 23 days, from May 7 to May 30, 1924, of 120 pounds is surprising when it is considered that tank No. 25, which contained the same kind of soil, but which was bare and uncultivated, lost only 57 pounds of water in a period of over four years. The loss from the plants in 23 days was more than twice as much as from the bare soil in four years.

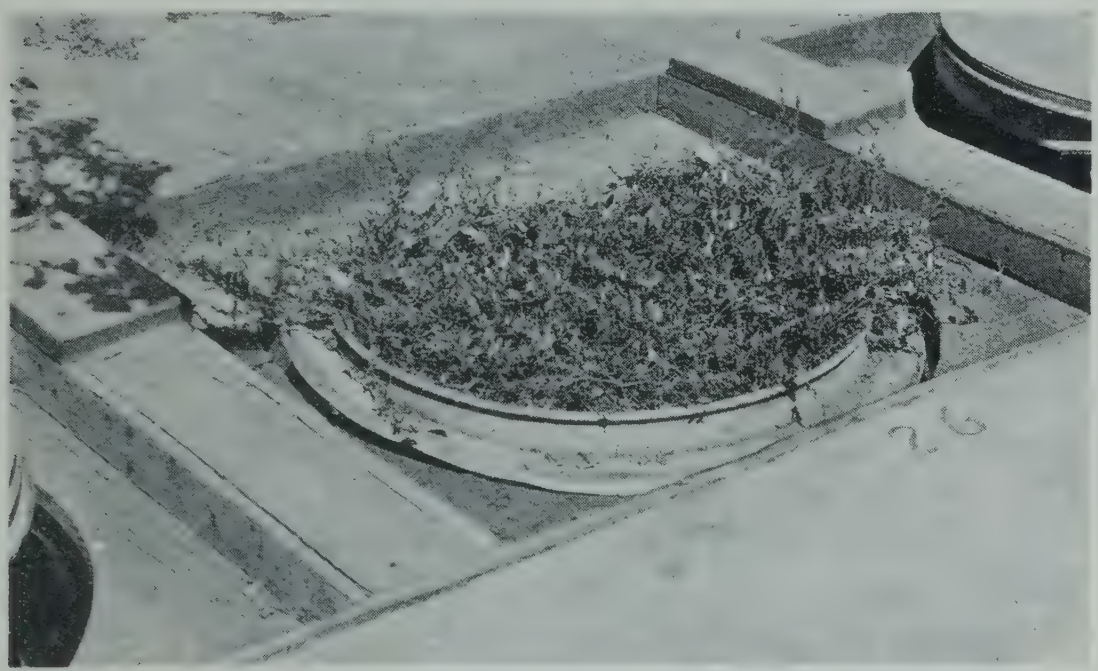


Fig. 35. Tank 26, containing morning glory (*Convolvulus arvensis*) plants. Photographed August 19, 1924.

A number of such data might be cited, all of which would show that the amount of water lost by direct evaporation from the surface of the soil under conditions prevalent in California is extremely small when compared to that transpired by plants.

That the loss of moisture by evaporation from the surface of the soil is a very small portion of the total losses from the soil may be true only under conditions governing these experiments. The losses of moisture by evaporation may become a large portion of the total amount of water applied to the soil, whether by rainfall or by irrigation, if these applications are in small amounts and are made on light soils in the warmer part of the year when evaporation is high.

Small amounts of water applied will wet the soil to shallow depths only and evaporation will cause much of the moisture retained in these depths to be lost. However, in California the rains usually come when the atmospheric evaporation power is low, and the usual practice is to apply water in irrigation in amounts sufficient to wet the soil to considerable depths. Therefore, the amount lost from the upper few inches of soil is a small portion of the total amount applied.

The Loss of Moisture by Evaporation from Bare Soils in Field Plots

Trials with field plots also were made to determine the loss of moisture by direct evaporation from the soil. The effect of cultivation was studied in these trials as it was in the tanks. The plots were located at Davis, on Yolo loam soil; at Mountain View, on Yolo clay loam soil; at Delhi, in the San Joaquin Valley, on Oakley fine sand; and at Whittier on a Yolo clay. These locations represent a wide range in climatic conditions and in soils. At each place four plots were laid out and prepared for irrigation. Each of the plots was 20 feet wide by 90 feet long, an area large enough to be cultivated by farm machinery, and yet not too large to be adequately sampled for moisture determinations. In all but one of these trials, that at Whittier, the plots were arranged side by side. The arrangement of the plots is shown in figure 36. The plots were separated from each other by a dry strip 10 feet wide. The places where samples were taken are indicated by the circles and numbers. The plots at Whittier were arranged in pairs, one uncultivated plot being opposite one cultivated plot.

Fairly level pieces of ground were selected, far enough removed from trees to prevent the extraction of moisture through their roots. Levees were constructed to enclose the four plots, and several cross-levees were thrown up across them to enable a more even application of the water. The water applied to each plot through the delivery ditch to the ditches along the high sides of each plot was measured over a weir with a hook-gage. It was possible to apply a definite depth of water to each plot by this means. Samples were taken before irrigation and the water necessary to wet the soil to a depth of five feet was calculated and applied.

Two of the plots at each location were cultivated to a depth of six inches as soon after irrigation as was possible. The cultivations were repeated at weekly intervals until the end of the experiment.

The other two plots were untouched, except to remove weeds by careful hoeing, as they appeared. Different types of tools were used in the different locations to cultivate the plots. Usually a disk was used first and then a spring-tooth harrow. In fact, the cultivations given each week were done as thoroughly as possible, and were more thorough than those given by the average orchardist.

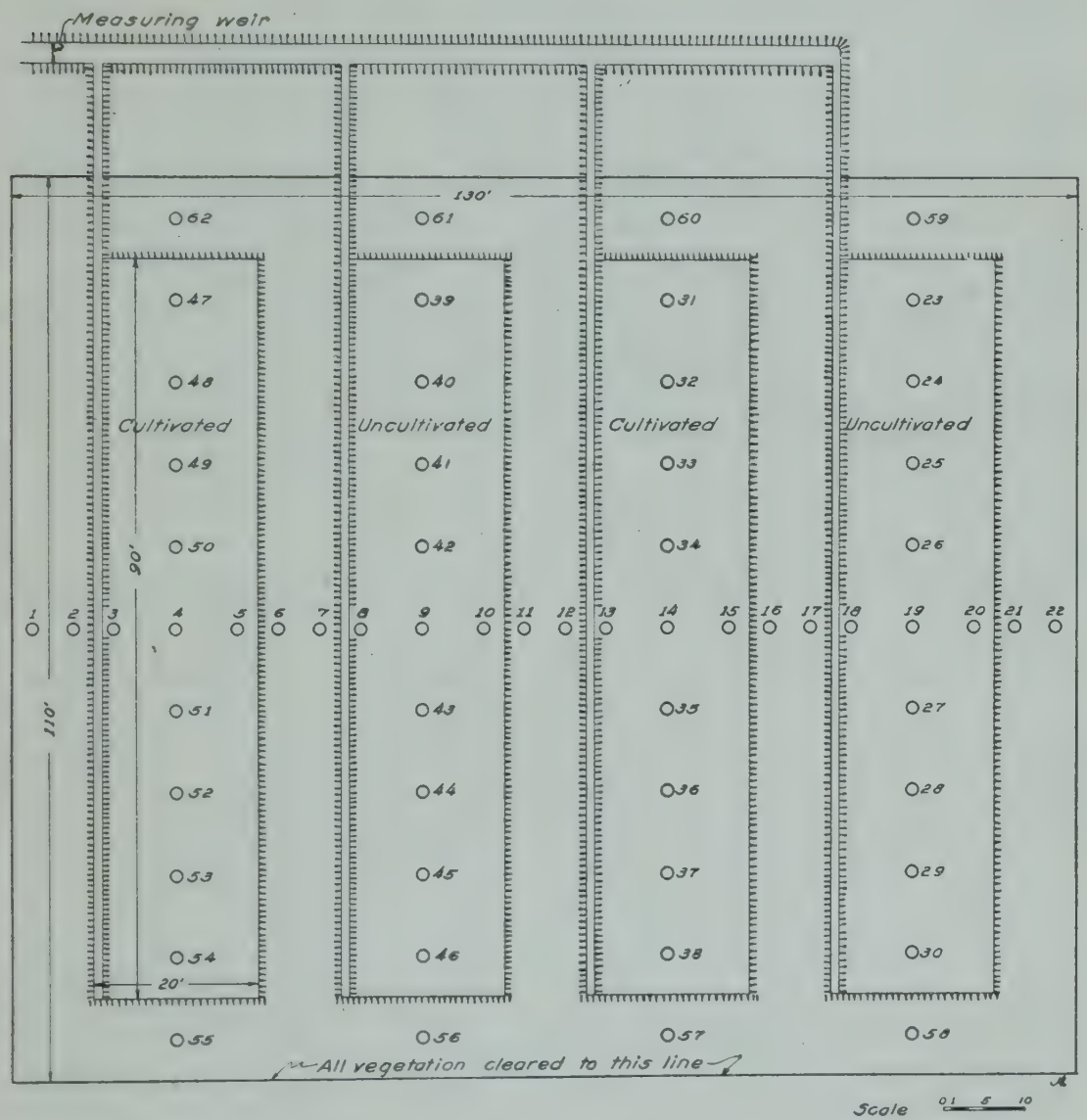


Fig. 36. The arrangement of the field plots used to determine the loss of moisture from bare cultivated and bare uncultivated soils. The places where samples were taken are indicated by circles and numbers.

The condition of the soil two months after irrigation in the cultivated plots at Whittier is shown in figure 37, and the condition of the uncultivated plots is shown in figure 38. These conditions were typical of those at the other locations. The cultivated plots were cultivated immediately after taking the weekly set of samples for moisture determinations. In the uncultivated plots there was some

Fig. 37. Condition of the Yolo clay soil in the cultivated plots at Whittier, two months after irrigation.

Fig. 38. Condition of the uncultivated Yolo clay soil in the uncultivated plots at Whittier, two months after irrigation. The place of sampling is indicated by the stake in the foregound and the special tube used to take samples in 4-inch depths is shown.

disturbance of the soil at the places where samples were taken, but it may be seen from figure 38 that this was slight and certainly not equivalent to cultivation.

Method of Sampling the Plots

Samples were taken with a soil tube, and all of the soil removed from the hole was retained in making moisture determinations. The soil from the upper 3 feet was placed in a soil can, and the soil from the second 3 feet, or from 3 to 6 feet, was kept separately and moisture contents were determined for these two depths of soil. Samples

Fig. 39. The manner of spacing the holes around the stake indicating the place of sampling for moisture determinations in the field plots used in the evaporation trials. Photograph taken in one of the uncultivated plots at Mountain View two months after irrigation.

were also taken in depths of 4 inches to a total depth of 20 inches in all of the plots, and in some cases the soil-moisture content in foot depths was determined. The special large soil tube shown in figures 37 and 38 was used to take these 4-inch samples.

Eleven samples were taken in each plot as indicated in figure 36. These were spaced 10 feet apart, and the outer holes, such as 47, 54, 39, and 46, were 5 feet from the edge of the plots. Samples, which were only 2½ feet from the wetted areas, were taken in the strips between the plots as indicated by holes numbered 1, 2, 6, 7, 11, 12, etc. Samples, indicated by holes 55 to 62 in figure 36, were also taken around the outer edges of the plots.

The manner of sampling at each place is shown in figure 39, which indicates the condition in one of the uncultivated plots in the Santa Clara Valley at Mountain View, two months after irrigation. The first sample was taken at a definite distance from the stake which had been placed before irrigation. The hole made at the next sampling was a measured distance from the last hole. This method was followed until a circle was completed around the stake. Then the place of sampling was moved out 6 inches further from the stake and a new circle of holes was started. The holes made in sampling in the uncultivated plots were not refilled, since it was thought that this addition of soil might be considered to constitute a mulching. Of course, in the cultivated plots the stakes were removed before cultivating. These were replaced before the next set of samples were taken by surveying from fixed points outside the plots, so that the places of sampling were fixed. In this way the possibility of obtaining the same type of soil at each sampling is thought to have been greater than if even a much greater number of samples were taken at random in the plots.

Attention again is called to the difficulty in obtaining representative samples even in such small areas as the plots used in the present study. Considerable variation was found in the amount of moisture contained in different samples taken simultaneously from the same localities in the plots. Of course, this is due in part to differences in the water retentiveness of the soil in the samples and it is not improbable that errors were caused by inequalities in drying the samples. Drying usually extended over long periods and was never less than three days with the larger samples, but even then constant weights were not always obtained.

It must be kept in mind that the interpretation of soil-moisture data is extremely difficult, and wide differences must be obtained to be significant. However, the probable errors are listed in each case, since they do serve to indicate the range of variation of the individual values used in calculating the means. It is thought that the number of samples taken was sufficiently great to calculate the significance of the results by the standard method; that is, the difference must be 3.2 times the probable error of the difference before it begins to be significant.

The means of moisture equivalent determinations of the soils in the different plots are given in table 32. At least 250 samples from each group of plots at each of the five locations were centrifuged.

These moisture equivalent determinations were made on 30-gm. weighed samples and they may be used to indicate the relative moisture retentiveness of the different soils.

Results of Sampling the Plots

The data obtained from the plots at Davis are summarized in table 33. The moisture content of the plots was quite high before irrigation. The plots were irrigated with amounts of water estimated to raise the moisture content of the soil to 22 per cent to a depth of 6 feet. The water was applied on August 2, 1921, and the tests were run until September 23, 1921, after which rains caused them to be discontinued. In addition to the fact that no real difference exists between the moisture contents in the cultivated and uncultivated plots, the record of the moisture conditions in the soil from 3 to 6 feet in depth is especially interesting since the data indicate that there was no change in the moisture content below the third foot. The samples taken on August 3 were from 12 to 20 hours after irrigation. Those on August 5 were taken approximately 72 hours after irrigation. Probably there was a downward movement of moisture between the samplings on August 3 and August 5, but from August 5 to September 23 there was practically no loss from the lower 3 feet of soil.

TABLE 32

Mean Values for the Moisture Equivalents of the Soil in the Plots Used to Determine Losses by Evaporation

Location of plot	Soil	Samples of soil from 0–3 feet in depth	Samples of soil from 3–6 feet in depth	Average for 0–6 feet in depth
Delhi	Oakley fine sand	5.77±0.77	11.09±0.24	8.43±0.12
Davis	Yolo loam	23.82±0.14	20.77±0.19	22.29±0.12
Santa Clara	Yolo clay loam with gravel	15.02±0.14	20.13±0.27	17.57±0.15
Whittier	Yolo clay	24.18±0.07	26.65±0.10	25.41±0.06

The relatively slight losses of moisture from the soil below the first foot after the moisture had become distributed, a condition which was observed in all of these tests, are indicated in table 34. The moisture content of the soil in foot depths to a depth of 6 feet is given in this table, and it will be noted from these data that the losses of moisture from the soil seem to be confined almost entirely to the first foot.

In order to convey some idea of the relative quantities of water present in the soil at different times during the test, the moisture contents given in table 34 are given in table 35 in amounts of water in acre-inches to the acre.

The plots at Mountain View, in the Santa Clara Valley, were irrigated on June 26, 1921. The results of sampling the soil for the determination of the moisture contents are given in tables 36 and 37. The results of sampling in the cultivated and uncultivated plots are very close. As in the plots at Davis, it was found that the moisture content of the lower 3 feet of soil remained practically constant. The samples taken on June 27 were made from 12 to 24 hours after the water was applied; and some downward movement may have taken place between this time and that of the sampling on June 28, which was about 48 hours after irrigation.

TABLE 33

SUMMARY OF SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN PERCENTAGES ON A DRY-WEIGHT BASIS IN THE BARE CULTIVATED AND UNCULTIVATED PLOTS AT DAVIS. IRRIGATED AUGUST 2, 1921

Depth of soil sampled, feet		Aug. 3	Aug. 5	Aug. 8	Aug. 15	Aug. 26	Sept. 23
0.3	Cultivated	23.7	21.0	20.3	18.8	18.3	18.1
		±0.86	±0.32	±0.25	±0.26	±0.26	±0.19
	Uncultivated	25.5	22.3	21.1	19.7	19.5	18.0
		±0.57	±0.18	±0.22	±0.16	±0.17	±0.14
3–6	Cultivated	19.1	17.9	20.0	18.2	19.2	18.2
		±0.29	±0.52	±0.42	±0.32	±0.27	±0.31
	Uncultivated	20.5	19.4	18.9	18.5	19.3	18.6
		±0.65	±0.37	±0.25	±0.24	±0.25	±0.27
0–6	Cultivated	21.4	19.4	20.2	18.5	18.8	18.2
		±0.45	±0.30	±0.24	±0.21	±0.19	±0.18
	Uncultivated	23.0	20.8	20.0	19.1	19.4	18.3
		±0.43	±0.21	±0.17	±0.14	±0.15	±0.15

Barley had been planted on the areas where the plots subsequently were laid out at Mountain View and at Davis. The crops were removed just before the tests were started. The amounts of water transpired by the plants had not reduced the moisture contents of

the soils from the depth of 3 to 6 feet below that to which they had been raised by the winter rains. The amounts of water applied to the plots at the beginning of the tests were estimated to be sufficient to raise the moisture content of the soil to its field capacity to a depth of 6 feet, and in so far as could be judged by appearance and by feeling, the soil below the top 4 to 8 inches in the plots at both locations remained in this condition throughout the period of observation.

TABLE 34

SUMMARY OF SOIL-MOISTURE CONTENTS IN FOOT DEPTHS IN PERCENTAGES ON A DRY-WEIGHT BASIS IN THE BARE CULTIVATED AND UNCULTIVATED PLOTS AT DAVIS. IRRIGATED AUGUST 2, 1921

Depth of soil sampled, feet		Aug. 3	Aug. 5	Aug. 8	Aug. 15	Aug. 26	Sept. 23
0–1	Cultivated	25.9 ±0.79	22.2 ±0.37	20.2 ±0.65	16.3 ±0.63	15.3 ±0.53	16.2 ±0.52
	Uncultivated	27.4 ±0.52	23.4 ±0.28	20.0 ±0.22	17.4 ±0.20	17.9 ±0.28	15.2 ±0.26
1–2	Cultivated	22.6 ±0.86	21.1 ±1.01	21.0 ±0.69	20.6 ±0.36	19.8 ±0.32	19.9 ±0.32
	Uncultivated	25.9 ±0.35	23.6 ±0.25	21.3 ±0.21	21.3 ±0.37	21.1 ±0.31	20.0 ±0.23
2–3	Cultivated	21.7 ±1.04	20.4 ±0.94	19.9 ±0.74	20.6 ±0.54	20.0 ±0.35	21.6 ±0.84
	Uncultivated	23.3 ±1.14	21.9 ±0.34	21.9 ±0.25	20.8 ±0.33	21.7 ±0.38	19.5 ±0.45
3–4	Cultivated	18.4 ±0.61	16.9 ±0.64	18.5 ±0.86	17.0 ±0.45	17.7 ±0.32	17.1 ±0.26
	Uncultivated	20.5 ±0.70	19.4 ±0.74	17.8 ±0.57	17.9 ±0.54	18.1 ±0.36	18.2 ±0.36
4–5	Cultivated	18.7 ±0.53	17.2 ±0.97	19.2 ±1.29	17.7 ±0.82	18.6 ±0.59	17.9 ±0.51
	Uncultivated	19.3 ±0.87	20.1 ±0.59	19.6 ±0.44	18.2 ±0.69	19.4 ±0.84	19.3 ±0.59
5–6	Cultivated	20.7 ±0.44	20.7 ±0.99	20.3 ±1.11	18.0 ±1.31	21.0 ±0.88	20.6 ±0.55
	Uncultivated	22.2 ±0.87	21.3 ±0.65	21.9 ±0.58	21.8 ±0.63	22.8 ±1.06	22.4 ±0.83

TABLE 35

SUMMARY OF SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN ACRE-INCHES TO THE ACRE IN BARE CULTIVATED AND UNCULTIVATED PLOTS AT DAVIS. IRRIGATED AUGUST 2, 1921

Depth of soil sampled, feet		Aug. 3	Aug. 5	Aug. 8	Aug. 15	Aug. 26	Sept. 23
0–3	Cultivated	11.09	9.83	9.50	8.80	8.56	8.46
		±0.40	±0.15	±0.17	±0.12	±0.12	±0.09
	Uncultivated	11.94	10.44	9.88	9.22	9.13	8.42
		±0.27	±0.08	±0.10	±0.07	±0.08	±0.07
3–6	Cultivated	8.94	8.38	9.36	8.52	8.99	8.52
		±0.14	±0.24	±0.20	±0.15	±0.13	±0.15
	Uncultivated	9.59	9.08	8.85	8.66	9.03	8.70
		±0.30	±0.17	±0.12	±0.16	±0.12	±0.13
0–6	Cultivated	20.03	18.21	18.86	17.32	17.55	16.98
		±0.42	±0.28	±0.22	±0.20	±0.18	±0.17
	Uncultivated	21.53	19.52	18.73	17.88	18.16	17.12
		±0.40	±0.20	±0.16	±0.13	±0.14	±0.14

TABLE 36

SUMMARY OF SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN PERCENTAGES ON A DRY-WEIGHT BASIS IN THE BARE CULTIVATED AND UNCULTIVATED PLOTS AT MOUNTAIN VIEW, IN THE SANTA CLARA VALLEY IRRIGATED JUNE 26, 1921

Depth of soil sampled, feet		June 27	June 28	June 30	July 7	July 14	July 21	July 28	Aug. 4
0–3	Cultivated	12.7	11.3	10.4	9.8	9.4	9.5	9.3	8.1
		±0.51	±0.37	±0.36	±0.36	±0.29	±0.32	±0.26	±0.27
	Uncultivated	11.3	10.6	11.3	10.6	10.4	9.6	9.9	8.9
		±0.53	±0.44	±0.38	±0.34	±0.44	±0.29	±0.31	±0.29
3–6	Cultivated	16.2	14.3	13.9	15.3	15.9	15.9	15.5	14.9
		±0.54	±0.44	±0.43	±0.38	±0.41	±0.42	±0.40	±0.33
	Uncultivated	15.1	15.3	14.8	15.4	15.5	15.0	14.9	15.0
		±0.38	±0.42	±0.34	±0.26	±0.29	±0.26	±0.28	±0.29
0–6	Cultivated	14.5	12.8	12.2	12.1	12.7	12.7	12.4	11.5
		±0.37	±0.29	±0.28	±0.26	±0.25	±0.26	±0.24	±0.21
	Uncultivated	13.2	13.0	13.1	13.0	13.0	12.3	12.4	12.0
		±0.33	±0.30	±0.26	±0.21	±0.26	±0.19	±0.21	±0.20

The moisture equivalents for the soil in these Santa Clara Valley plots do not agree with the moisture content in the soil after irrigation. The soil was quite gravelly, as figure 39 shows. The field samples, of course, include much of the gravel which holds very little water, and the moisture content is calculated on the basis of the total dry weight of the soil in the samples. Therefore, as might be expected, the field moisture content determinations are lower than the moisture equivalents which are made only on soil particles less than 2 mm. in diameter.

TABLE 37

SUMMARY OF THE SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN ACRE INCHES TO THE ACRE IN THE CULTIVATED AND UNCULTIVATED PLOTS AT MOUNTAIN VIEW, IN THE SANTA CLARA VALLEY
IRRIGATED JUNE 26, 1921

Depth of soil sampled, feet		June 27	June 28	June 30	July 7	July 14	July 21	July 28	Aug. 4
0–3	Cultivated	6.43	5.73	5.27	4.97	4.76	4.81	4.71	4.10
		±0.26	±0.19	±0.18	±0.18	±0.15	±0.16	±0.13	±0.14
	Uncultivated	5.73	5.37	5.73	5.37	5.27	4.86	5.02	4.51
		±0.27	±0.22	±0.19	±0.17	±0.22	±0.15	±0.16	±0.15
3–6	Cultivated	8.21	7.25	7.04	7.75	8.06	8.06	7.85	7.55
		±0.27	±0.22	±0.22	±0.19	±0.21	±0.21	±0.20	±0.17
	Uncultivated	7.65	7.75	7.50	7.80	7.85	7.60	7.55	7.60
		±0.19	±0.21	±0.17	±0.13	±0.15	±0.13	±0.14	±0.15
0–6	Cultivated	14.64	12.98	12.31	12.72	12.82	12.87	12.56	11.65
		±0.37	±0.29	±0.28	±0.26	±0.25	±0.26	±0.24	±0.21
	Uncultivated	13.38	13.12	13.23	13.17	13.12	12.46	12.57	12.11
		±0.33	±0.30	±0.26	±0.21	±0.26	±0.19	±0.21	±0.20

The summary of the moisture determinations in the cultivated and uncultivated plots at Delhi are given in tables 38 and 39. Table 38 gives the moisture contents in percentages and table 39 gives the same data calculated as acre inches to the acre. These plots were irrigated on July 7, 1921, and the soil was wet to a depth of 5½ feet. The determinations of moisture content were carried on until December 3, 1921, after which heavy rains caused the sampling to be discontinued. The rainfall from September 9 to December 3 was 0.76 inches. This came in light showers; most of it evaporated soon after the surface of the soil was wetted. The soil was wetted by these rains only to a depth of about 4 inches.

The test at Delhi was repeated during 1922, the same plots being used as in 1921. The results of sampling the plots during 1922 are given in table 40. The plots were not irrigated in 1922 but were wet by rains which did not cease until early in May. No further rain fell until after the last date of sampling, September 8. A rainfall of 0.48 inches occurred just before the first set of samples was taken on May 9, 1922, and the soil was found to be wet to the full depth of 6 feet on this date.

TABLE 38

SUMMARY OF SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN PERCENTAGES ON A DRY-WEIGHT BASIS IN THE BARE CULTIVATED AND UNCULTIVATED PLOTS AT DELHI, 1921. IRRIGATED JULY 7, 1921

Depth fo soil sampled, feet		July 8	July 9	July 11	July 16	July 23	July 30	Aug. 6	Aug. 20	Sept. 9	Dec. 3
0–3	Cultivated.....	7.6	6.6	6.1	5.7	5.4	4.8	4.6	4.2	3.7	4.6
		±0.19	±0.24	±0.17	±0.12	±0.09	±0.11	±0.07	±0.07	±0.05	±0.07
	Uncultivated....	8.1	6.8	6.1	5.2	4.8	4.4	4.0	3.5	3.3	4.3
		±0.32	±0.21	±0.16	±0.10	±0.07	±0.08	±0.05	±0.05	±0.06	±0.03
3–6	Cultivated.....	6.5	6.2	6.2	7.2	7.5	7.4	7.6	7.1	7.7	7.1
		±0.32	±0.24	±0.32	±0.34	±0.28	±0.42	±0.32	±0.29	±0.27	±0.20
	Uncultivated....	6.3	5.9	7.4	6.3	7.1	7.4	7.0	7.3	6.4	7.7
		±0.29	±0.22	±0.26	±0.32	±0.29	±0.26	±0.28	±0.24	±0.30	±0.24
0–6	Cultivated.....	7.1	6.4	6.2	6.5	6.5	6.1	6.1	5.6	5.7	5.9
		±0.18	±0.17	±0.17	±0.18	±0.15	±0.22	±0.16	±0.15	±0.14	±0.11
	Uncultivated....	7.2	6.4	6.8	5.8	6.0	5.9	5.5	5.4	4.9	6.0
		±0.21	±0.15	±0.15	±0.20	±0.15	±0.14	±0.14	±0.12	±0.16	±0.12

The cultivated plots were given the same treatment in 1922 as in 1921, and an attempt was made to keep the uncultivated plots free from weeds. At Delhi, as well as at the other localities, this was found to be very difficult during the first week or two after the wetting of the soil. The weeds could not be removed or scraped off with a hoe without disturbing the surface of the soil until they were several inches high and their presence could be detected. The loss of moisture through transpiration even from such small plants may have been appreciable under the hot interior valley conditions in California, and the apparent difference in soil-moisture content of about ½ of 1 per cent between the cultivated and uncultivated plots at Delhi from July 16 to the last date of sampling in 1921 may be due, at least in part, to this cause.

The rapidity with which water was taken from the sandy soil at Delhi by the rather sparse grasses is illustrated by the following experiment: A series of samples was taken on April 15 from an area

immediately adjoining the plots on which some grasses had been allowed to grow during the spring of 1922. The soil-moisture content in the upper 3 feet was 6 per cent and 11 per cent in the lower 3 feet. Samples taken May 9, showed that the moisture content of the upper 3 feet of soil was 2.7 per cent and that of the depth from 3 to 6 feet was 5.0 per cent. On June 30 the average moisture content of the upper 3 feet of soil was only 0.8 per cent, and samples could not be taken below the 3-foot depth since the dry sand would fall into the hole made by the soil tube.

TABLE 39

SUMMARY OF SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN ACRE INCHES TO THE ACRE IN THE CULTIVATED AND UNCULTIVATED PLOTS AT DELHI, 1921. IRRIGATED JULY 7, 1921

Depth of soil sampled, feet		July 8	July 9	July 11	July 16	July 23	July 30	Aug. 6	Aug. 20	Sept. 9	Dec. 3
0–3	Cultivated.....	3.94	3.42	3.17	2.96	2.80	2.48	2.39	2.18	1.92	2.39
		±0.10	±0.12	±0.09	±0.06	±0.05	±0.06	±0.04	±0.04	±0.03	±0.04
	Uncultivated....	4.20	3.53	3.16	2.70	2.49	2.28	2.07	1.82	1.71	2.23
		±0.16	±0.12	±0.08	±0.05	±0.04	±0.04	±0.03	±0.03	±0.03	±0.02
3–6	Cultivated.....	3.37	3.21	3.22	3.74	3.89	3.84	3.94	3.68	3.99	3.68
		±0.17	±0.12	±0.17	±0.18	±0.14	±0.22	±0.17	±0.15	±0.46	±0.10
	Uncultivated....	3.26	3.06	3.84	3.27	3.68	3.84	3.63	3.78	3.32	3.99
		±0.15	±0.11	±0.14	±0.17	±0.15	±0.14	±0.14	±0.12	±0.15	±0.12
0–6	Cultivated.....	7.31	6.63	6.39	6.70	6.69	6.32	6.33	5.82	5.91	6.07
		±0.19	±0.18	±0.18	±0.18	±0.16	±0.23	±0.17	±0.16	±0.10	±0.11
	Uncultivated....	7.46	6.59	7.00	5.97	6.17	6.12	5.70	5.60	5.03	6.22
		±0.22	±0.16	±0.16	±0.21	±0.16	±0.15	±0.15	±0.12	±0.17	±0.12

The soil below 3 feet at Delhi is very fine, compacted and cemented, but the water slowly penetrates into this layer. The larger probable errors in the averages of moisture contents of samples taken from 3 to 6 feet in these plots indicates the variability of this soil. On the other hand, the smaller probable errors in the means of the results of sampling from 0 to 3 feet indicate that this layer is fairly uniform. The average moisture equivalent of the soil in the cultivated plots in the upper 3 feet was found to be 5.98 per cent and that of the second 3 feet was 11.78 per cent, while the moisture equivalents of the two depths of soil in the uncultivated plots were 5.49 per cent and 10.40 per cent, respectively. This indicates that the soil in the uncultivated plots had a smaller water-holding capacity than that in the cultivated plots. This also is suggested by the results of the sampling on May 9, 1922, reported in table 40.

The total losses of moisture by evaporation seem to be greater from the sandy soil than from the finer textured soils. However, these losses seem to be confined almost entirely to the upper layers of soil. The data in tables 38, 39, and 40 indicate that no losses occurred from the 3 to 6 foot depth.

TABLE 40

SUMMARY OF SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN PERCENTAGES ON A DRY-WEIGHT BASIS IN THE CULTIVATED AND UNCULTIVATED PLOTS AT DELHI, 1922

Depth of soil sampled, feet		May 9	June 30	July 24	Sept. 8
0–3	Cultivated	6.3±0.03	4.0±0.04	3.5±0.04	2.9±0.04
	Uncultivated	5.5±0.06	3.3±0.06	2.9±0.05	2.5±0.04
3–6	Cultivated	12.0±0.29	10.8±0.37	10.6±0.31	10.6±0.31
	Uncultivated	10.8±0.43	9.4±0.27	9.3±0.36	9.8±0.40
0–6	Cultivated	9.1±0.15	7.4±0.19	7.0±0.16	6.7±0.16
	Uncultivated	8.1±0.21	6.4±0.14	6.1±0.18	6.2±0.20

The plots at Whittier on the clay soil were irrigated July 15, 1921. A depth of water equivalent to 4 inches was applied to all of the plots. Although a crop of barley had been raised during the previous winter and spring, the soil seemed still to be moist on July 14, 1921, when samples were taken before applying the water. On this date, the average moisture content of the soil from 0 to 3 feet was 11.8 per cent and from 3 to 6 feet it was 12.6 per cent. After the water was applied, the surface of the soil remained so wet that the first set of samples following irrigation could not be taken until July 21.

The results of the sampling in the plots at Whittier, given in table 41, seem low when compared with the moisture equivalents for these plots given in table 32, but the soil seemed to be amply moist. This high moisture content was apparent when the samples taken in the areas where the water had been applied were compared with those taken in the strip between the plots. This lack of agreement between the maximum field capacity and the moisture equivelant, in this case, may be due to the difficulty in making moisture equivalent determinations on heavy soils when 30-gram samples are used as had been pointed out by Joseph and Martin[35] and Veihmeyer, Israelsen and Conrad.[51]

The percentages of moisture given in table 41 have been calculated as equivalent amounts of water in acre-inches to the acre and are reported in table 42. Here, again, it will be seen that the differences between the moisture contents of the soil in the cultivated and uncultivated plots are not significant and that the losses of moisture are confined to the upper 3 feet of soil, since the data show no change in moisture content in the lower 3 feet of soil throughout the period of observation.

TABLE 41

SUMMARY OF THE SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN PERCENTAGE ON A DRY-WEIGHT BASIS IN THE CULTIVATED AND UNCULTIVATED PLOTS AT WHITTIER. IRRIGATED JULY 15, 1921

Depth of soil sampled, feet		July 21	July 28	Aug. 4	Aug. 11	Aug. 18	Sept. 3	Sept. 20
0–3	Cultivated	17.3 ±0.25	16.0 ±0.20	16.2 ±0.26	16.2 ±0.21	15.6 ±0.18	16.4 ±0.22	15.9 ±0.18
	Uncultivated	18.0 ±0.24	17.1 ±0.18	16.4 ±0.15	16.5 ±0.13	15.7 ±0.18	16.4 ±0.15	15.5 ±0.15
3–6	Cultivated	15.3 ±0.21	15.1 ±0.22	15.3 ±0.26	15.5 ±0.28	14.8 ±0.22	15.1 ±0.21	15.0 ±0.24
	Uncultivated	14.3 ±0.27	14.4 ±0.32	14.3 ±0.32	14.5 ±0.31	14.2 ±0.30	14.4 ±0.29	14.5 ±0.31
0–6	Cultivated	16.3 ±0.16	15.6 ±0.15	15.8 ±0.18	15.9 ±0.17	15.2 ±0.14	15.7 ±0.15	15.5 ±0.15
	Uncultivated	16.2 ±0.18	15.8 ±0.15	15.4 ±0.18	15.5 ±0.17	15.0 ±0.18	15.4 ±0.17	15.0 ±0.19

That the losses of moisture by evaporation directly from the surface of the clay soil were so small is surprising. It was thought that this soil would crack badly and thus the moisture content of the soil in the uncultivated plots would be lower than that in the cultivated plots. Apparently the uncultivated soil did not crack enough to influence the results. The amount of cracking in the uncultivated plots can be noted in figure 38.

It is a surprising fact that in the field trials, as well as in the studies of evaporation from bare soils in tanks, the uncultivated soils did not crack to an appreciable extent. However, the Yolo clay, the Yolo clay loam, and the Yolo loam cracked badly when they were dried by the extraction of moisture by plants growing on them. The absence of cracking as well as the data reported in the tables indicate

that the loss of moisture from the bare soils was slight. However, in this connection, it must be remembered that soils of certain types of high colloidal content crack badly even though plants are not allowed to grow on them, and even though they do not dry out appreciably.

Approximately two months after the water was applied samples were taken in all of the plots to determine the distribution of moisture in the upper layers of soil. The moisture contents of the soil in 4-inch layers to a total depth of 20 inches were obtained from samples taken with a soil tube of large diameter (fig. 38), and the results are reported in table 43.

TABLE 42

SUMMARY OF THE SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN ACRE INCHES TO THE ACRE IN THE CULTIVATED AND UNCULTIVATED PLOTS AT WHITTIER. IRRIGATED JULY 15, 1921

Depth of soil sampled, feet		July 21	July 28	Aug. 4	Aug. 11	Aug. 18	Sept. 3	Sept. 20
0–3	Cultivated	9.59	8.87	8.98	8.98	8.55	9.09	8.82
		±0.14	±0.11	±0.14	±0.12	±0.10	±0.12	±0.10
	Uncultivated	9.98	9.54	9.09	9.15	8.71	9.09	8.60
		±0.13	±0.10	±0.08	±0.07	±0.10	±0.13	±0.11
3–6	Cultivated	8.49	8.38	8.54	8.60	8.21	8.38	8.32
		±0.12	±0.12	±0.14	±0.15	±0.12	±0.12	±0.13
	Uncultivated	7.93	7.99	7.93	8.05	7.88	7.99	8.05
		±0.15	±0.18	±0.18	±0.17	±0.17	±0.16	±0.17
0–6	Cultivated	18.04	17.25	17.52	17.58	16.76	17.47	17.14
		±0.18	±0.17	±0.20	±0.19	±0.16	±0.17	±0.17
	Uncultivated	17.91	17.53	17.12	17.20	16.59	17.08	16.65
		±0.20	±0.17	±0.20	±0.19	±0.20	±0.19	±0.20

These data show clearly that the losses of moisture by evaporation were confined largely to the upper 8 inches of soil, and that the greater portion of the loss from this depth of soil occurred in the first 4 inches. When the moisture contents given in table 43 are compared with those for the upper 3 feet of soil given in tables 33 to 42, it seems that there were slight losses at least to a depth of 20 inches. The results obtained in the study of the losses by evaporation from bare soils in tanks also show that water was lost throughout the entire depth of the soil (table 31). However, the loss below the surface layer was at an extremely slow rate and would be negligible in

amount for even longer periods than the usual ones between irrigations or between rains. It can not be assumed that those losses below the surface layers are due entirely either to upward movement of moisture by capillarity or to loss by water vapor movement. Amounts of water lost from the soil by water vapor are generally held to be very small, the work of Buckingham[17] and Bouyoucos[7] frequently being cited to show that this is true. However Bouyoucos and McCool[10] have recently pointed out that considerable aeration of soils takes place because of atmospheric pressure changes, and the slight losses of moisture from the deeper layers of soils in the plots and in the tanks may have been due to movement of air and water vapor through them. Of course, the ease of air movement between the soil mass and the sides of the tanks was greater than that for movement through the soil in the field plots, and it should be expected that moisture would be lost throughout a greater depth of soil in the tanks.

TABLE 43

SUMMARY OF MOISTURE CONTENTS IN FOUR-INCH DEPTHS OF SOIL IN PERCENTAGES ON A DRY-WEIGHT BASIS IN CULTIVATED AND UNCULTIVATED PLOTS IN 1921. SAMPLES TAKEN APPROXIMATELY TWO MONTHS AFTER IRRIGATION

Location of plots	Treatment	Depths of soil samples in inches				
		0 to 4	4 to 8	8 to 12	12 to 16	16 to 20
Davis	Cultivated	6.6 ±0.37	15.7 ±0.51	20.1 ±0.47	19.1 ±0.28	18.6 ±0.23
	Uncultivated	8.6 ±0.39	15.5 ±0.23	19.0 ±0.41	19.0 ±0.21	19.4 ±0.21
Mountain View	Cultivated	4.0 ±0.20	9.5 ±0.41	10.2 ±0.34	10.3 ±0.37	10.7 ±0.41
	Uncultivated	3.9 ±0.15	9.1 ±0.28	10.4 ±0.36	11.0 ±0.67	10.9 ±0.79
Delhi	Cultivated	1.3 ±0.06	3.9 ±0.12	4.1 ±0.12	4.1 ±0.10	4.2 ±0.15
	Uncultivated	1.5 ±0.05	3.1 ±0.06	3.3 ±0.06	3.7 ±0.09	4.1 ±0.25
Whittier	Cultivated	4.1 ±0.22	11.5 ±0.33	15.1 ±0.54	16.2 ±0.31	15.9 ±0.23
	Uncultivated	4.1 ±0.21	11.0 ±0.28	16.2 ±0.43	17.3 ±0.39	16.5 ±0.47

It should be noted from the data in table 43 that the only significant differences in moisture content of the soil in 4-inch depths in the cultivated and uncultivated plots appears to be in the 4 to 8 and 8 to 12 inch depths in the plots at Delhi. The difference in moisture content in favor of the cultivated plots is 0.8 ± 0.13 per cent in each instance. This is equivalent in amount to a depth of water of only 0.09 inches in the 8 inches of this sandy soil. In every other case, the differences were insignificant, and it is apparent that thorough cultivation at weekly intervals for a period of approximately two months failed to result in a saving of moisture or to influence its movement by capillarity. In fact, the results reported in tables 33 to 43, as well as those secured in the studies of the loss of moisture from bare soils in tanks indicate that the movement of water by capillarity from moist soils to drier soils is extremely slow and is effective only through very short distances.

TABLE 44

SUMMARY OF SOIL-MOISTURE CONTENTS IN THREE-FOOT DEPTHS IN PERCENTAGES ON A DRY-WEIGHT BASIS IN SAMPLES TAKEN FROM THE UNWETTED AREAS BETWEEN AND AROUND THE CULTIVATED AND UNCULTIVATED PLOTS AT THE BEGINNING AND AT THE END OF THE TESTS

Location of plots	Depth of soil samples	Moisture content at beginning of tests	Moisture content at end of tests
Davis	0–3	14.6±0.55	14.9±0.40
	3–6	17.2±0.46	17.3±0.29
Mountain View	0–3	6.7±0.23	6.0±0.22
	3–6	13.1±0.28	13.2±0.27
Delhi	0–3	1.7±0.14	1.3±0.09
	3–6	6.0±0.93	6.2±0.57
Whittier	0–3	12.3±0.25	12.0±0.22
	3–6	15.1±0.43	14.6±0.45

This is further substantiated by the records of the moisture contents of the samples taken in the strips between the plots and those taken outside the wetted areas around the plots (see figure 36) which are reported in table 44. There are no significant differences in the moisture contents of the samples taken at the beginning and of those taken at the end of the tests. Some of these samples were taken 5 feet from the wetted areas but the majority were taken only $2\frac{1}{2}$ feet

away. The latter samples, taken at frequent intervals, showed no increase in moisture content throughout the duration of the experiments. This indicates that there probably was no movement of moisture from one plot to another and also that there was little if any loss of moisture from the plots by lateral movement. Certainly the lateral movement to a distance of only 2½ feet was not enough to change the moisture content at this point. The results of further studies on the movement of moisture from moist soils to drier soils are given in the following pages.

The Movement of Moisture from Moist Soils to Drier Soils in Columns

Since the data from the studies of the losses of moisture from bare soils in tanks and in field plots indicate that the movement of moisture from moist soils to drier soils, when the source of water is not a free water surface, is slight in amount and extent, an effort was made to secure more direct evidence concerning such movement by studying the behavior of moisture in columns of soils. The columns used in this study are the same as those described by McLaughlin.[40*]

The columns were made of redwood planks 2 inches thick and 8 inches wide, nailed together and lined with galvanized iron, and were therefore 6 inches by 6 inches inside and 6 feet long.

All of the columns were packed with Yolo clay loam taken from the surface of the orchard at the Deciduous Fruit Station of the University of California at Mountain View. The gravel was screened and the soil was thoroughly mixed. Midway from end to end the columns were packed with air-dry soil, this being held in place by tight-fitting boards at both ends of the soil section. In filling this central section the soil was added in uneven thin layers, then compackted evenly. The weight of water-free soil was known and the column was frequently weighed during the process of packing so that a definite volume weight of soil corresponding to that of the soil in place in the field was obtained. Weighed quantities of water were then added to raise the soil to its field capacity. The soil was then covered and allowed to stand for 48 hours, after which samples were taken and the moisture content determined. If the water seemed to be uniformly distributed throughout the soil mass, the holes made in sampling were refilled with soil properly moistened, and this soil

* The writer is indebted to Mr. W. W. McLaughlin of the Division of Agricultural Engineering of the Bureau of Public Roads, United States Department of Agriculture, for the use of this equipment.

was tamped into place. The supports for the pieces of boards holding the central section of wetted soil in place were removed and the end sections packed with the drier soils. The packing was done in the same manner as that for the central section, and the same volume weight was obtained. The end boards which retained the central section now were removed and the spaces occupied by them were thoroughly packed with the drier soil. Strips of asphaltic roofing paper were laid along the edges of the planks and a plate glass cover was securely clamped into place and the columns placed upright.

Since it was recognized there were mechanical difficulties in securing good capillary contact of the moist with the drier soil, and since poor contact might inhibit movement from the moist to the drier soil, great care was taken to avoid, as much as possible, error due to this cause. At the termination of the period of observation with each column and after the final samples were taken, the column was cut lengthwise and the place where the moist and drier soils were originally joined was carefully noted. There was no evidence of discontinuity of the soil masses in any case and the original line of demarcation between moist and drier soil could not be detected.

It was very difficult to raise the moisture content of the rather large quantities of drier soils required for the end sections to the percentages required. Although there were some departures from the moisture contents it was desired to bring about in the end sections of the columns a fairly satisfactory method was finally used. The dry soil was placed in a large metal can and water in sufficient quantity to raise it to the necessary percentage was added by spraying it on the soil in a fine mist, meanwhile rotating the can so that the soil was constantly mixed.

The amounts of water contained in the central and end sections of the columns at the beginning of the tests are not so accurately known as is the distribution of moisture at the end. Samples were taken from the ends and near the middle of each section before the columns were placed upright but only a few samples could be taken at the beginning of the test without too much disturbance of the soil. Variations of 0.7 per cent in the samples from the drier soil were obtained, while the samples from the moist soil showed a variation of about 1 per cent. The moisture contents of the soil when packed are probably accurate to within 1 per cent. The moisture equivalents made on the samples taken from the packed columns varied from 21 to 24 per cent and averaged about 22 per cent, an average which may be taken as being fairly close to the field capacity for this soil.

Several types of small soil augers were tried in taking the samples both at the beginning and at the end of the tests, but a small soil tube seemed to give the best results; at least the moisture percentages obtained with this device were higher than those with the augers, and the results secured seemed to be consistent.

The moisture contents given in the following figures were obtained from the columns at the end of the tests. The columns were taken down and the glass plates removed. The samples were taken along the center line of the column but were staggered so that one sample would not interfere with another. In taking the sample, the soil tube was pushed the full depth of the column. The moisture contents recorded, then, are the average amounts of moisture contained in the depth of the column. No attempt was made to determine the distribution of moisture in this depth. It should be noted that the movement of moisture in both an upward and downward direction determined by noting the change in color of the drier soil in every case was always less than the movement indicated by the results of sampling the soil. This might be accounted for if there were a greater movement of moisture down the back of the columns than down the front, which was covered with plate glass, and this may account for the discrepancy in the two methods of indicating the movement of moisture.

In this connection it must be mentioned that the soil columns were subject to fluctuations in temperature. Temperature variations may have caused a greater movement in one portion of the soil mass than in another. Furthermore, condensation of moisture on the sides of the container due to temperature changes and subsequent downward movement of this condensed water may account for some of the downward movement in the wet section of the columns.

The determination of the extent and rate of movement by noting the change in color of the drier soil was extremely difficult and practically impossible in the columns in which the end sections were packed with soils having the higher moisture contents. It is believed that the distribution of moisture as indicated by sampling the soil is a much more reliable and accurate measure of the extent of moisture movement than by noting the change in color of the drier soil, especially in the clay loam soils used in the present studies.

The columns selected, the data from which are presented here, are typical of all of the columns studied and represent the range in moisture contents used. Figure 40 graphically illustrates the distribution of moisture in one of the 6-foot columns, the central 2-foot

section of which was packed with a soil containing 22 per cent of moisture. The upper section was packed with a soil of 3 per cent moisture content, and the bottom section with soil containing 3.5 per cent of moisture. This column was started on August 26, 1922, and the samples were taken on January 17, 1923. The results of the sampling indicate the extent and distribution of moisture after the soils had been in contact for 144 days.

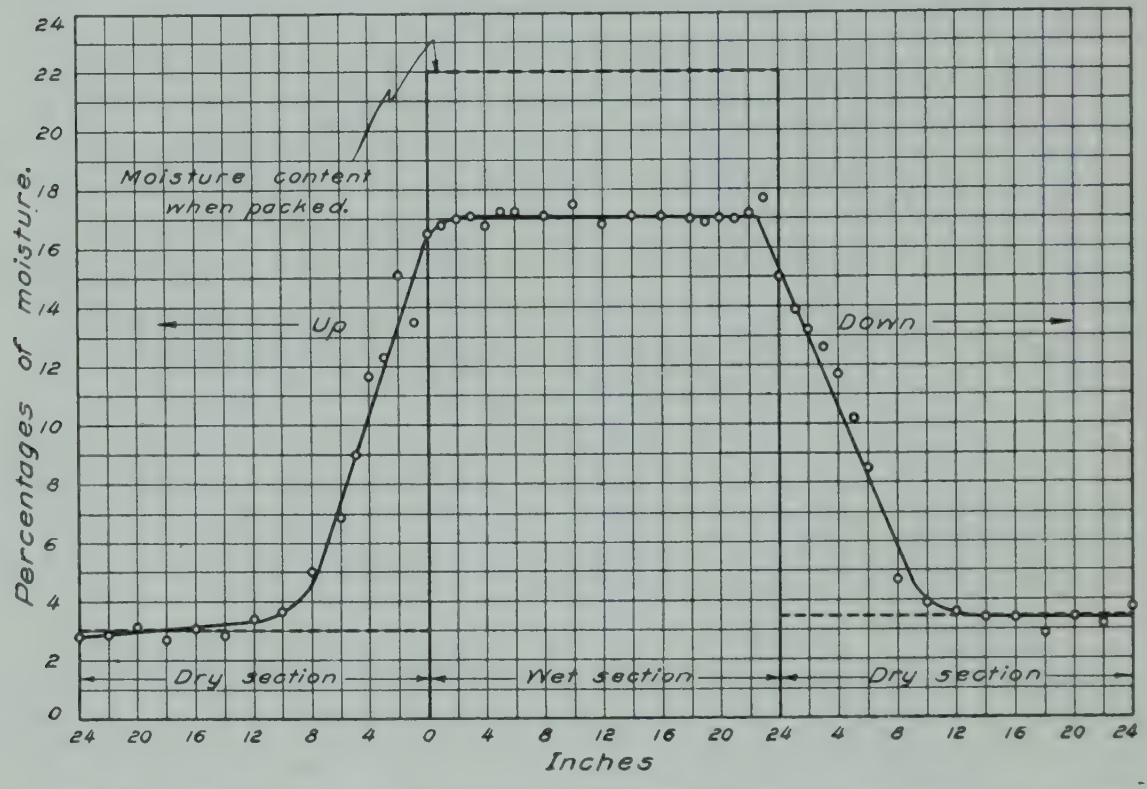


Fig. 40. The movement of moisture upward and downward (left and right in diagram) from soil mass initially containing 22 per cent of moisture to soil containing 3 per cent and 3.5 per cent of moisture. Column was started August 26, 1922, and samples were taken January 17, 1923. The place of sampling in the column and the amount of moisture found are indicated by the circles. Depth is shown by numerals along the base of diagram.

The upward movement of moisture from the moist into the dry soil in this column, as measured by the change in color of the dry soil, was 3.0 inches and the downward movement was 5.75 inches. It is evident that both of these distances are less than the movement indicated by the results of sampling, which also showed no difference in extent of moisture movement upward and downward. The results obtained by sampling in all of the columns showed this to be the case in every instance. However, McLaughlin,[40] using the same equipment, reports a greater downward movement in each of his tests. Attempts were made to determine the rate of movement of moisture from the moist soil into the drier soil by noting the advance of the

moist layer. These were not entirely successful owing to inability to determine clearly the line of demarcation between moist and drier soil. However, it was clear that the greater portion of the movement took place within the first few days after the soils were placed in contact. McLaughlin[40] concludes from his studies "that the greater part of capillary distribution of the water occurs while water is being applied and in the next two or three days thereafter."

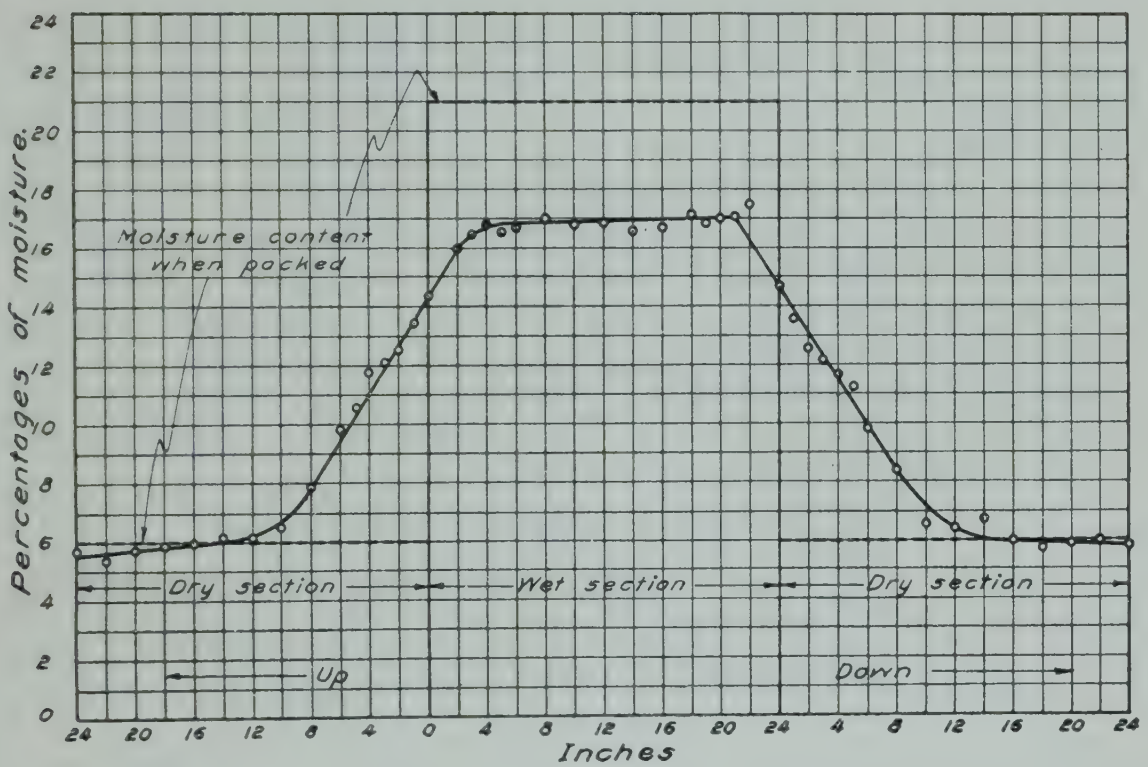


Fig. 41. The movement of moisture upward and downward (left and right in diagram) from soil mass initially containing 21 per cent of moisture to soils containing 6 per cent of moisture. Column was started August 29, 1922, and samples were taken January 17, 1923. The place of sampling in the column and the amount of moisture found are indicated by the circles. Depth is shown by numerals along the base of diagram.

The distribution of moisture and the extent of upward and downward movement from a soil with an initial moisture content of 21 per cent, occupying the central section of one of the columns, into soils with an initial moisture content of 6 per cent, in the end sections are shown in figure 41. The test with this column was started on August 29, 1922, and the samples were taken on January 17, 1923, after a period of 141 days. The upward movement of moisture from the moist soil into the drier soil was 5.4 inches, and the movement downward was 5.1 inches, as indicated by the change in color of the drier soil.

Figure 42 illustrates the distribution of moisture and the extent of movement in a column the central section of which was packed with soil which was wet to a water content of 23 per cent. The upper end section was packed with soil at 7.8 per cent and the lower end section with soil having an initial moisture content of 8 per cent. The test was continued for 141 days, from August 29, 1922, to January 17, 1923. The measurement of the line of demarcation

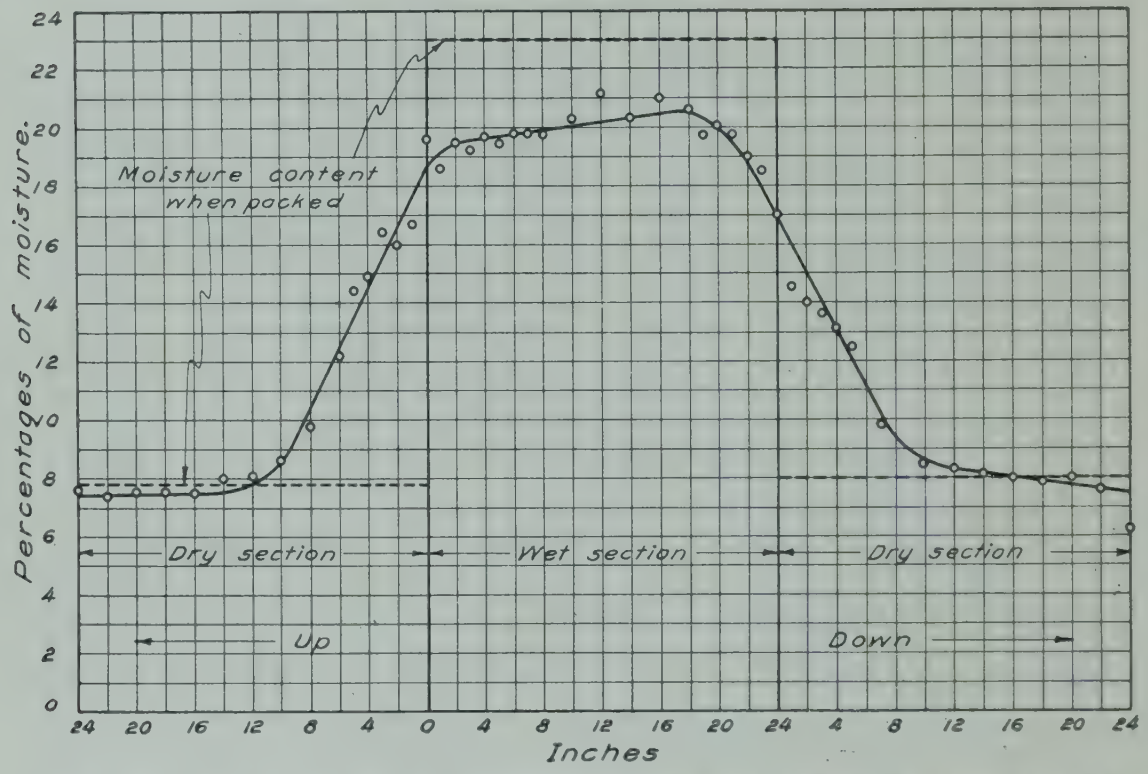


Fig. 42. The movement of moisture upward and downward (left and right in diagram) from soil mass initially containing 23 per cent of moisture to soils containing 7.8 and 8 per cent of moisture. The column was started August 29, 1922, and samples were taken January 17, 1923. The place of sampling in the column and the amount of moisture found are indicated by circles. Depth is shown by numerals along the base of diagram.

between moist and drier soil at the end of the test, made by noting the change of color, indicated that the upward movement was 3.25 inches and the downward movement was 6.7 inches. The results presented in figure 42 indicate the distribution and extent of moisture movement from soil initially wet to about its field capacity into soil approximately at the hygroscopic coefficient.

The results of sampling a column of soil the central section of which was packed with soil and wetted so that the moisture content at the start was 21.5 per cent, and the end sections were packed with soil containing 12.5 per cent of moisture, are illustrated in figure 43. The soils were in contact for 140 days. Thus the end sections initially had a moisture content approximately equal to the calculated wilting

coefficient for this soil, and the movement of moisture indicated is from a soil at about the field capacity into one at the wilting coefficient. It was very difficult, because of the relatively high moisture content, to distinguish any change in the color of the drier soil used in this column as the moisture moved. The best estimate which could be made by this method was a movement upward of 3.25 inches and downward of 3.3 inches.

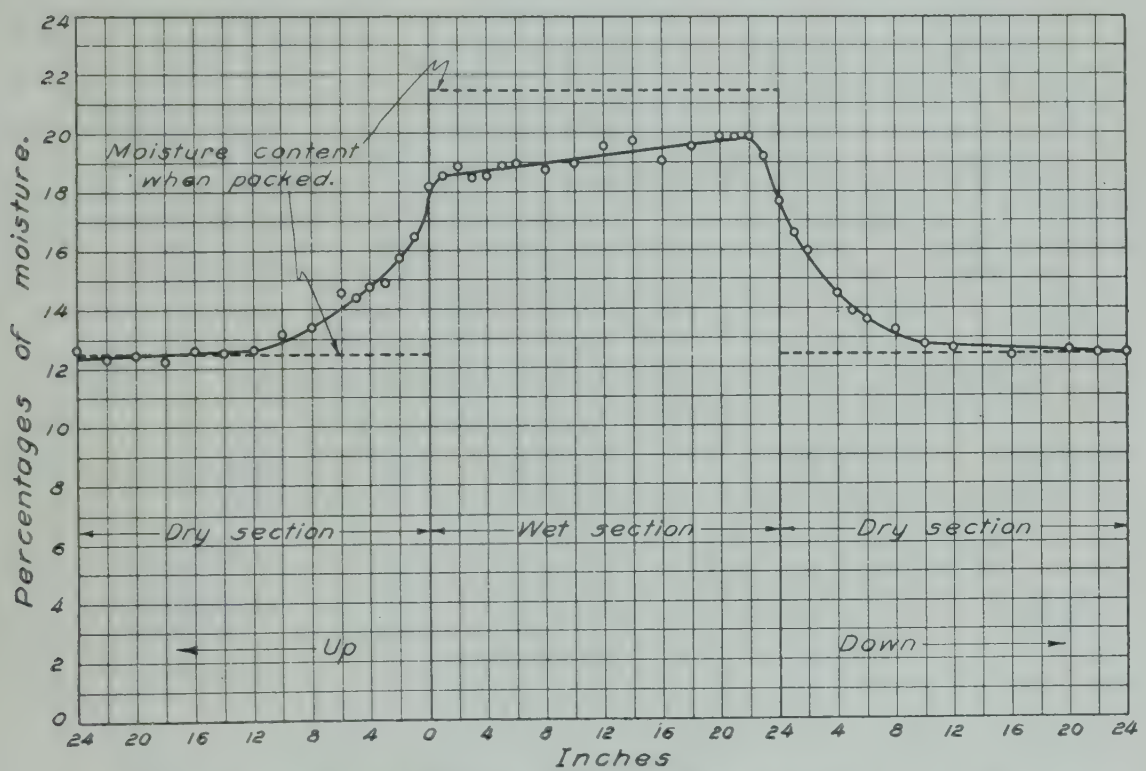


Fig. 43. Movement of moisture upward and downward (left and right in diagram) from soil mass initially containing 21.5 per cent of moisture to soil containing 12.5 per cent of moisture. Column was started September 1, 1922, and samples were taken January 18, 1923. The place of sampling in the column and the amount of moisture found are indicated by the circles. Depth is shown by numerals along the base of diagram.

The distribution and extent of moisture movement from a soil wetted to 22 per cent into soil with an initial moisture content of 14.5 per cent is shown in figure 44. The initial moisture content of the soil in the end sections of this column was as high as could be used in these experiments and yet be materially less than the field capacity. It was necessary to stir the soil constantly while applying the water, in order to bring it to an intermediate moisture content. Puddling would occur in soils at moisture contents higher than 14.5 per cent. These soils were in contact for 139 days—from September 2, 1922, to January 18, 1923. The movement upward as indicated by the color change in the drier soil was 3.2 inches and the downward movement was 3.9 inches, but these measurements were very indefinite.

The data selected for presentation here were taken from columns covering a range of moisture contents in the drier soils from an air-dried condition to a moisture content above the wilting coefficient, and they indicate the extent of moisture movement into these drier soils from a clay loam soil wet to its field capacity.

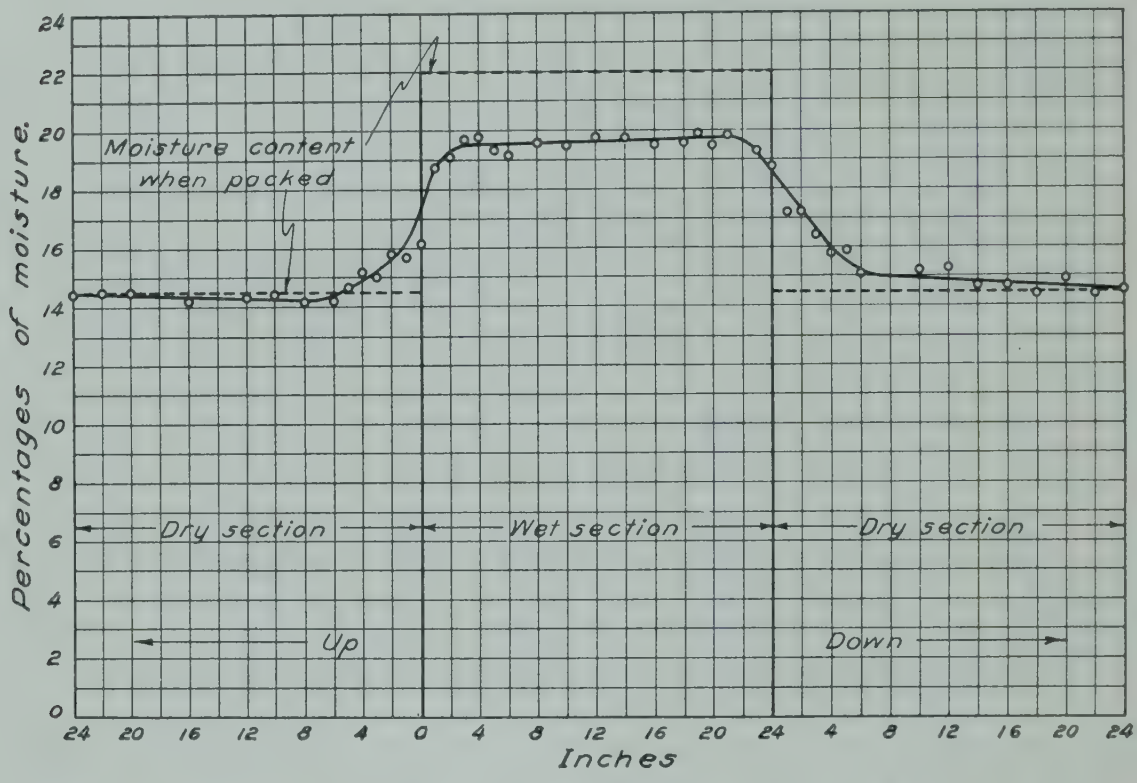

Percentages of moisture.
Moisture content when packed
Dry section
Wet section
Dry section
Up
Down
Inches
24 22 20 18 16 14 12 10 8 6 4 2 0
24 20 16 12 8 4 0 4 8 12 16 20 24 4 8 12 16 20 24

Fig. 44. Movement of moisture upward and downward (left to right in diagram) from soil mass initially containing 22 per cent of moisture to soil containing 14.5 per cent of moisture. Column started September 2, 1922, and samples taken January 18, 1923. The place of sampling in the column and the amount of moisture found are indicated by the circles. Depth is shown by numerals along the base of diagram.

McLaughlin[40] has suggested that the rate and extent of movement of moisture varies with the initial percentage of moisture in the wet pack. Therefore, it is probable that the extent of moisture movement shown in these figures is greater than that under field conditions wherein the moisture content of the wet mass of soil is rapidly being depleted by plants growing on it. Apparently the moisture content of the drier soil had little effect upon the extent of movement of moisture from the moist to drier soil.

It is evident that in the clay loam soil used in these columns the movement of moisture from the moist to the drier soil in either direction, even during long periods of time, was limited. The results of these, as well as the other studies herein reported, indicate that the capillary movement of moisture from a moist soil to a drier soil,

when the soil is not in contact with a free water-surface, is too limited in extent and probably in rate to be effective for use by plants.

In view of the data presented herein concerning the capillary movement of moisture, the prevalent belief as to the results of light and heavy irrigations is plainly incorrect. The belief that the moisture content of all of the soil occupied by the roots of the trees will be raised to a certain percentage by the application of small amounts of water, because of the downward capillary movement of the water with a consequent equalization of the moisture content of all of the soil, is not in accord with results obtained in these experiments. A light irrigation, in fact, results in wetting the soil to a less depth than the application of a larger amount.

Summary

The records of moisture conditions in mature prune orchards in the Santa Clara Valley of California show that the soil-moisture supply is constantly changing during the growing season. The maintenance of a uniform soil-moisture content, a condition often specified as essential for the best fruit production, probably is impossible. An approximation of a uniform soil-moisture content possibly might be brought about by very frequent applications of water. However, this practice would be objectionable not only for reasons of labor and expense involved and the interference with other orchard practices, but also because of the probable injurious effects on the soil.

The amount of water that may be stored in the soil at one application is limited. The records of soil-moisture conditions taken from the upper 6 feet of soil indicate that the moisture supply usually is exhausted within four or five weeks in a loam soil in a mature orchard during the growing season.

Irrigation during the dormant season for the purpose of storing water for use by the trees during the growing season usually is ineffective in years of normal rainfall in such localities as the Santa Clara Valley. In a year of normal rainfall, the upper 6 feet of soil in the Santa Clara Valley are usually filled with water to the maximum field or capillary capacity at the beginning of the growing season.

While a considerable amount of water was taken by the roots of the trees from the soil below 6 feet, it was much less than that taken from the upper 6 feet of soil. Also, the rate of extraction of moisture was much lower from the lower depths of soil than from

the upper 6 feet. Wilting was always noted when the moisture content of the upper 6 feet of soil had been reduced to the wilting coefficient. Therefore, the moisture content of the upper 6 feet of soil probably exerts a much greater influence on such trees than that of the soil below 6 feet. However, the moisture in these lower depths of soil probably does maintain trees during long periods after the moisture supply of the upper 6 feet has been exhausted.

The parallelism of the graphs representing the rate of loss of moisture from the upper 3 feet of soil and that from the next lower 3 feet of soil indicates that the roots of the prune trees, under the conditions of these observations, are uniformly distributed in the upper 6 feet of soil. The parallelism of these graphs also indicates that probably no movement of moisture takes place between the top 3 feet and the next 3 feet of a soil during the growing season in the absence of free water.

The moisture content of the upper 6 feet of soil in these prune orchards was reduced to, or below, the wilting coefficient toward the end of the growing season. This condition usually persisted for two or more months before the moisture supply was replenished. In every case the inability of the roots of the trees to obtain water from soil below 6 feet at a sufficiently rapid rate was evidenced by wilting and shedding of leaves.

The soil-moisture records presented here indicate that the use of water by the mature prune trees does not seem to be influenced by the amount of water present in the soil, provided the soil-moisture content has not been reduced below the wilting coefficient. The slope of the graphs representing the moisture conditions apparently are substantially the same, whether the soil-moisture supply be high or low. The intensity of atmospheric evaporating power and leaf area seemed to determine the use of water by the trees; the amount of available water present in the soil, and the condition or state of growth of the trees, except in so far as this affected leaf area, seemed to be of secondary importance.

The yields, drying ratios, size, and quality of prunes produced did not seem to be related to the frequency or amounts of water applied to the soil. The yields, drying ratios, size and quality of Muir peaches were not influenced by the amounts or times of irrigation, except in years of unusually low rainfall or when the soil-moisture content had previously been reduced below the wilting coefficient. Mature prune and peach trees did not seem to be affected by changes in soil-moisture content unless the moisture content of the upper 6 feet of soil had been reduced to about the wilting coefficient.

The results obtained in the studies of the Muir peaches were in general the same as those obtained with the mature prune trees. It was possible to make more detailed measurements of the growth of the peach trees. These indicated that, with the exception of the season of 1920, during which the soil moisture in the unirrigated rows was depleted early in the season, there was no difference in growth made which could be related to differences in soil-moisture conditions in the different rows.

Studies of young prune trees grown in tanks under controlled conditions indicate that the use of water by these young trees was not influenced by the amount of water in the soil above the wilting coefficient. Under comparable atmospheric conditions the rate of extraction of moisture by the roots of the trees was the same whether the moisture content of the soil above the wilting coefficient was high or low. Apparently the roots of these trees were able to obtain water as readily when the soil moisture content had been reduced almost to the wilting coefficient as when the soil was filled with water to its maximum field capacity.

Because of the comparatively slow capillary movement of moisture, serious objections may be raised to previous water relation studies wherein dependence has been placed upon capillary movement to cause a uniform distribution in the soil of water applied at any point. A predetermined soil-moisture content, less than the full field capacity, could not be brought about in the large masses of soils used in these experiments. It is also very probable from the results obtained that even if a relatively low moisture content could be established uniformly throughout the soil mass, a condition which probably is impossible of attainment, the moisture content would very quickly be reduced by the growing plant and the relatively low moisture content could not be maintained under natural field conditions or even in controlled experiments.

The results obtained from the controlled studies made with prune trees in tanks indicate that not only the use of water but the trees themselves were not affected by variations in amounts of soil moisture above the wilting coefficient. While these results apply only to these young prune trees, it appears that many of the current views regarding soil-moisture relations of other plants may also be questioned.

When the atmospheric evaporating power was judged, by means of the measurements and apparatus employed, to be the same in the spring as in the fall, the use of water to a unit of leaf area apparently was the same. This suggests that the use of water to a unit of leaf area by these young prune trees was not influenced by the state of

growth. Within the limits of weighings made in these experiments, it was not possible to detect differences in the use of water to a unit of leaf area during the fore part of the growing season, when the trees were making rapid length growth, and the use of water in the latter part of the season, when length growth had ceased and the leaves were more mature. The intensity of the atmospheric evaporating power and leaf-area seemed to govern the use of water.

When the moisture content of the loam soils on which these young prune trees were grown was reduced below a rather definite amount, the trees wilted and did not recover until water was added to the soil. The wilting coefficient, which was calculated from the moisture equivalent, was very close to the actual moisture content found in the soil at the beginning of permanent wilting. This agreement between the theoretical wilting coefficient and the residual moisture found in the soil at the time when the trees permanently wilted persisted throughout several seasons and at different times during each season the observations were made. This indicates that, within the range of conditions under which these experiments were made, atmospheric conditions had little influence upon the amount of residual moisture in the loam soils at the beginning of permanent wilting.

While the wilting coefficient of the soils under observation in these experiments was a percentage of soil moisture at which the young trees grown in tanks, as well as the mature prune trees, began to wilt permanently, the soil moisture was reduced much below this percentage. However, the soil-moisture supply was reduced to the hygroscopic coefficient in only a few cases and only by mature prune trees. Usually about one-half of the soil-moisture between the wilting coefficient and the hygroscopic coefficient was taken by the trees. Vetch plants grown in tanks were able to reduce the moisture content of the soil only to a like amount. Although the wilting coefficient, which is a critical soil-moisture content, is not the lower limit of available moisture, it probably is a better basis for comparing moisture properties of soils than the hygroscopic coefficient.

The time of fall coloration and abscission of leaves of the young prune trees grown in tanks was the same when the soil-moisture content was high as when the soil-moisture content was low but not below the wilting coefficient. However, defoliation caused by wilting could be induced by withholding water until the soil-moisture content was reduced below the wilting coefficient.

It is probable that the beginning of dormancy or of maturity of the plant tissues is not affected by variations of soil-moisture content within a rather wide range. No injury from irrigation late in the

season, shortly before the leaves dropped, could be detected in the Santa Clara Valley prune orchards or the Muir peach orchard at Davis. This suggests that the hardiness of the wood of these mature trees and probably of the young prune trees in containers had not been affected by a high soil-moisture content late in the season.

Under conditions at Mountain View, in the Santa Clara Valley of California, where these observations were made, young prune trees grown in tanks lost a very slight amount of water by evaporation from the bare twigs and branches. The use of water by these young trees during the winter was so small that the need for the application of water to meet the current demands of the trees during this season is probably negligible, except in years of unusually light rainfall.

The losses of moisture by direct evaporation from the surfaces of soils in containers were measured by repeated weighings. Losses of moisture by direct evaporation from the soil surfaces of field plots were measured by sampling the soil and ascertaining the soil-moisture contents. A rather wide range of soils and climatic conditions was obtained in the field trials. These losses were found to be relatively slight in amount. A comparison of the evaporation losses from the surfaces of bare soils with the amount of water taken from the soil by plants showed that evaporation losses were extremely small portions of the total amounts of water lost from the soil.

The loss of moisture by direct evaporation from the soil was confined very largely to shallow depths of soil. Moisture below the upper eight inches of soil was lost at an extremely slow rate. The losses of moisture from the surfaces of soils exposed to evaporation for much longer periods of time than are usual between irrigations, were insufficient to prohibit the growth to maturity of barley and vetch plants. After exposure to evaporation for the entire summer, there was sufficient moisture in the soil in tanks below the upper eight inches to grow vetch plants to maturity without applying additional water.

Cultivation did not influence the losses of moisture by evaporation from the bare surfaces of the soils in the tanks and in the field plots under observation in these experiments. Cultivation did not materially influence the distribution of moisture in these soils. There were no significant differences in the moisture contents in 4-inch depths in the cultivated and uncultivated soils with the exception of the 4 to 8 and 8 to 12-inch depths at Delhi. The differences in these depths of this sandy soil were very small but were probably significant.

The loss of moisture by evaporation from the surfaces of soils immediately following the application of water were found to be a large portion of the total evaporation loss for a long period of time. About half of the loss in 80 days occurred in the first week after irrigation, and the greater amount of this was lost before the soil was in condition to be properly cultivated.

After the water applied to the soil had become distributed, the movement of moisture by capillarity was found to be extremely slow. There was neither upward nor downward movement in the soil from the 3-foot depth to the 6-foot depth during the time the observations were made in these experiments. The movement of moisture laterally in 2½ months was not sufficient to affect the moisture content of the soil 2½ feet from the wetted area.

The movement of moisture from moist soils to dry soils packed in columns and remaining in contact with each other for 4½ months was slight in amount and in extent in both an upward and downward direction. The results of these studies indicate that the capillary movement of moisture from moist soil to drier soil, when the soil is not in contact with a free water surface, is too limited in extent and probably in rate to be effective for the use by plants.

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LITERATURE CITED

[1] ALWAY, F. V.
1913. Studies on the relation of the non-available water of the soil to the hydroscopic coefficient. Nebraska Agr. Exp. Sta. Research Bul. **3**: 1–121

[2] ALWAY, F. V.
1918. Some studies in the dry land regions. U. S. Dept Agr., Bur. Pl. Ind. Bul. **130**: 1–212.

[3] ALWAY, F. V., and G. R. MCDOLE.
1917. Relation of movement of water in a soil to its hygroscopicity and initial moistness. Jour. Agr. Res., **10**: 391–428.

[4] BAKER, P. B.
1912. The moisture content of field soils under different treatments. Nebraska Agr. Exp. Sta., 35th Ann. Rept., **1911**: 106–111.

[5] BATCHELOR, L. D., and H. S. REED.
1919. Winter drying or die-back of the walnut. California Agr. Exp. Sta. Circ. **216**: 1–20.

[6] BATCHELOR, L. D., and H. S. REED.
1923. The seasonal variation of the soil moisture in a walnut grove in relation to the hygroscopic coefficient. California Agr. Exp. Sta. Tech. Paper **10**: 1–31.

[7] BOUYOUCOS, G. J.
1915. Effect of temperature on movement of water vapor and capillary moisture in soils. Jour. Agr. Res., **8**: 141–172.

[8] BOUYOUCOS, G. J.
1918. Relation between the unfree water and the heat of wetting of soils and its significance. Michigan Agr. Exp. Sta. Tech. Bul. **42**: 1–21.

[9] BOUYOUCOS, G. J.
1921. A new classification of soil moisture. Soil Sci., **11**: 33–47.

[10] BOUYOUCOS, G. J., and M. M. MCCOOL.
1924. The aeration of soils as influenced by air-barometric pressure changes. Soil Sci., **18**: 52–63.

[11] BRIGGS, L. J.
1897. The mechanics of soil moisture. U. S. Dept. Agr., Div. Soils, Bul. **10**: 1–24.

[12] BRIGGS, L. J., and H. L. SHANTZ.
1912. The wilting coefficient for different plants and its indirect determination. U. S. Dept. Agr., Bur. Pl. Ind. Bul. **230**: 1–83.

[13] BRIGGS, L. J., and H. L. SHANTZ.
1914. Relative water requirements of plants. Jour. Agr. Res., **3**: 1–63.

[14] Briggs, L. J., and H. L. Shantz.
1917. Comparison of the hourly evaporation rate of atmometers and free water surfaces with the transpiration rate of Medicago sativa. Jour. Agr. Res., **9**: 277–292.

[15] Briggs, L. J., C. A. Jensen, and J. W. McLane.
1917. The mulched-basin system of irrigated citrus culture and its bearing on the control of mottle-leaf. U. S. Dept. Agr. Bul. **499**: 1–32.

[16] Burr, W. W.
1914. The storage and use of soil moisture. Nebraska Agr. Exp. Sta., Research Bul. **5**: 1–88.

[17] Buckingham, E.
1907. Studies on the movement of soil moisture. U. S. Dept. Agr., Bur. Soils Bul. **38**: 1–61.

[18] Caldwell, J. R.
1913. The relation of environmental factors to the phenomena of permanent wilting. Physiological Researches, **1**: 1–56.

[19] Call, L. E., and M. C. Sewell.
1917. The soil mulch. Jour. Am. Soc. Agron., **9**: 49–61.

[20] Cameron, F. K.
1911. The soil solution. Chemical Pub. Co., Easton, Pa., 1–136.

[21] Chilcott, E. C., and J. S. Cole.
1918. Subsoiling, deep tilling, and soil dynamiting in the Great Plains. Jour. Agr. Res., **14**: 481–521.

[22] Copeland, E. B.
1898. A new self-registering transpiration machine. Bot. Gaz., **26**: 343–348.

[23] Corbett, L. C.
1898. An improved auxanometer and some of its uses. West Virginia Agr. Exp. Sta., 12th Ann. Rept., 40–42.

[24] Curtis, O. F.
1925. Studies on the tissues concerned in the transfer of solutes in plants. Ann. Bot., **39**: 573–585.

[25] Denny, F. E.
1922. Formulas for calculating number of fruits required for adequate sample for analysis. Bot. Gaz., **73**: 44–57.

[26] Fortier, S., and S. H. Beckett.
1912. Evaporation from irrigated soils. U. S. Dept. Agr., Off. Exp. Sta., Bul. **248**: 1–77.

[27] Fowler, L. W., and C. B. Lipman.
1917. Optimum moisture conditions for young lemon trees on a loam soil. Univ. Calif. Publ. Agr. Sci., **3**: 25–36.

[28] Gardner, W.
1919. Capillary moisture holding capacity. Soil Sci., **7**: 319–324.

[29] Hall, A. D.
1910. The soil. E. P. Dutton and Co., New York, 1–311.

[30] Harris, F. S., and H. W. Turpin.
1917. Movement and distribution of moisture in soil. Jour. Agr. Res., **10**: 113–153.

[31] Hilgard, E. W., and R. H. Loughridge.
1897. Endurance of drought in soils of the arid regions. California Agr. Exp. Sta. Rept., 1897–1898, pp. 40–64.

[32] Hoff, E. J.
1916. Sensitive water-level recorder. Engin. News, **76**: 974–975.

[33] Hoagland, D. R.
1925. Physiological aspects of soil solution investigations. Hilgardia, **1**: 228–257.

[34] Israelsen, O. W., and F. L. West.
1922. Water holding capacity of irrigated soils. Utah Agr. Exp. Sta., Bul. **183**: 1–24.

[35] Joseph, A. F., and F. J. Martin.
1923. The moisture equivalent of heavy soils. Jour. Agr. Sci., **13**: 49–57.

[36] Kiesselbach, T. A.
1915. Transpiration as a factor in crop production. Nebraska Agr. Exp. Sta. Bul. **6**: 1–214.

[37] King, F. H.
1914. Soil management. Orange Judd Company, New York, 1–303.

[38] Livingston, B. E., and Koketsu.
1920. The water supplying power of the soil as related to the wilting of plants. Soil Sci., **9**: 469–485.

[39] Loughridge, R. H.
1908. Distribution of water in the soil in furrow irrigation. U. S. Dept. Agr., Off. Exp. Sta., Bul. **203**: 1–63.

[40] McLaughlin, W. W.
1920. Capillary movement of soil moisture. U. S. Dept. Agr., Bul. **835**: 1–69.

[41] Miller, E. C.
1916. Comparative study of root systems and leaf area of corn and sorghums. Jour. Agr. Res., **6**: 311–331.

[42] Parker, F. W.
1922. The classification of soil moisture. Soil Sci., **13**: 43–54.

[43] Puri, A. N.
1925. A critical study of the hygroscopic coefficient of soils. Soil Sci., **15**: 272–283.

[44] Rotmistrov, V. G.
1913. Sushshnost Zasukhi po Dannym Odesskago Opytnago Polia Odessa, Russia. Russion ed., 1911, 1–66. (English translation.) The nature of drought according to the evidence of the Odessa Experiment Station. Sta. M. of L. and A., Dept. of Agr., Odessa.

[45] Sewell, M. C.
1919. Tillage: A review of the literature. Jour. Am. Soc. Agron., **11**: 269–290.

[46] Sleight, R. B.
1917. Evaporation from the surfaces of water and river-bed materials. Jour. Agr. Res., **10**: 209–261.

[47] Shive, J. W., and B. E. Livingston.
1914. The relation of atmospheric evaporating power to soil-moisture content at permanent wilting of plants. Plant World, **17**: 81–121.

[48] Shull, C. A.
1916. Measurement of the surface forces in soils. Bot. Gaz., **62**: 1–29.

[49] Thomas, M. D.
1921. Aqueous vapor pressure of soils. Soil Sci., **11**: 409–434.

[50] Tufts, W. P.
1919. Pruning of young deciduous fruit trees. California Agr. Exp. Sta., Bul. **313**: 1–37.

[51] Veihmeyer, F. J., O. W. Israelsen, and J. P. Conrad.
1924. The moisture equivalent as influenced by the amount of soil used in its determination. California Agr. Exp. Sta., Tech. Paper **16**: 1–62.

[52] Weaver, J. E., F. C. Jean, and J. W. Crist.
1922. Development and activities of roots of crop plants. Carnegie Inst. Wash., Pub. **316**: 1–117.

[53] Widtsoe, J. A.
1914. Principles of irrigation practice. The MacMillan Company, New York, 1–496.

[54] Willard, R. E., and E. P. Humbert.
1914. Soil moisture. New Mexico Agr. Exp. Sta. Bul. **86**: 1–86.

[55] Young, H. J.
1911. Soil mulch. Nebraska Agr. Exp. Sta. Rept., p. 124.

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PLATES 1-3

PLATE 1

Fig. 1. The effect of differences in soil moisture on the condition of prune trees. Photographed October 18, 1922. Tree 3 on the left, and tree 5 on the right on soil below the wilting coefficient. Tree 4 in the center on soil with moisture content near the wilting coefficient.

Fig. 2. The same trees shown in fig. 1, ten days later.

Fig. 1

Fig. 2

PLATE 2

Fig. 1. The effect of soil moisture on the condition of prune trees in the fall. Photographed October 7, 1922. Tree 15, on the left, and tree 17, on the right, on soil with moisture content below the wilting coefficient. Tree 16, in the center, on soil with moisture content near the wilting coefficient.

Fig. 2. Tree 19, in the center, and tree 20, on the right, on soil kept continuously above 16 per cent moisture content. Tree 18, on the left, on soil kept above 16 per cent moisture content until the last week in August when it was allowed to wilt but was revived, and the soil-moisture content thereafter allowed to fluctuate between the maximum field capacity and the wilting coefficient. At the time this photograph was taken, October 18, 1922, tree 18 was on soil near the wilting coefficient.

Fig. 1

Fig. 2

PLATE 3

Fig. 1. Fall condition of prune trees on water-logged soil, compared to that of a tree on soil near the wilting coefficient. The soil on which tree 12, on the left, and tree 14, on the right, were growing was water-logged throughout the growing season. Tree 13, in the center, was irrigated only when the soil-moisture content was reduced nearly to the wilting coefficient. At the time this photograph was taken, October 18, 1922, the moisture content of the soil on which tree 13 was growing was near the wilting coefficient.

Fig. 2. Prune trees in tanks used to determine the loss of moisture by evaporation from bare twigs and branches during the dormant season. Tree 1 is on the right, tree 3 is in the center, and tree 6 is on the left.

Fig. 1

Fig. 2

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ETIOLOGY AND TRANSMISSION OF ENDOSEPSIS (INTERNAL ROT) OF THE FRUIT OF THE FIG[1]

PANOS D. CALDIS[2]

FIGS, CAPRIFIGS, AND CAPRIFICATION

A comprehensive discussion of the diseases of the fruit of the fig should take into consideration its structural peculiarities. Eisen(10), (11) discusses exhaustively the morphology and structure of this fruit. Condit(5) gives a brief discussion of the fig fruit and its structure.

The fig is a nearly closed, more or less hollow receptacle, the inner walls of which are lined by the flowers when immature and by the fruit when ripe. It is not, therefore, a fruit in the strict botanical sense of the word but an aggregation of fruits lining the cavity of a hollow receptacle, technically a synconium, with an opening at the center of the flattened, distal end, which is closed during the early stages by a system of overlapping bracts. As the fig begins to ripen and soften these bracts or scales loosen and an opening is formed, the diameter of which varies from 2 to 10 mm., according to the variety of fig. This opening is usually referred to as the "eye" of the fig. In the ripe fig the wall of the receptacle to which the flowers are attached is called "the meat," and the aggregation of mature florets "the pulp." Figure 1 shows the internal appearance of the receptacle when split longitudinally. The flowers of the fig vary with the variety, the sex, and the crop.

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The fig is a dioecious, insect-pollinated plant, the staminate and pistillate flowers being borne on different trees. The male tree is known as the caprifig and in its receptacles we find chiefly two types of flowers. The staminate flowers are arranged, in most varieties, around the eye and the gall flowers (modified female flowers) occupy the rest of the cavity. The gall flowers have a very short style, particularly suited to the needs of the pollinating insect as will be explained later. The caprifig bears a succession of fruit known as the "mamme" or overwintering crop, the "profichi" or spring crop, which is the important one in relation to the edible figs, and the "mammoni" or fall crop. This succession of crops provides the proper habitat for the insects throughout the year. The caprifigs are not edible. They are usually rather small and non-succulent, the size varying with the variety. The names of the varieties of caprifigs most commonly grown in California and which were used in the experiments to be outlined later are: Roeding No. 1, Roeding No. 2, Roeding No. 3, Roeding No. 4, Markarian No. 1, Markarian No. 2, the Stanford, the Milco, and a number of seedling male trees.

The female tree bears also a succession of crops, one on the wood of last year's growth and a second on the new wood. These are the edible figs. In most cases the second crop only is of commercial importance. The receptacle of the edible fig is lined by a single kind of flowers, the pistillate, which resemble the gall flowers of the caprifig except that the style is much longer. The edible fig varieties are of two types, the parthenocarpic, or varieties which develop fruit without pollination, and the varieties that require pollination (caprification). The important varieties of the first category, grown in California, are the White Adriatic, the Black Mission, and the Kadota (Dottato), while the varieties which require pollination are chiefly those called Calimyrna, Stanford, and San Pedro. The first two were introduced into California from Asia Minor, the first being the commercially famous "Lob Injir" of Smyrna.

The process of pollinating the pistillate flowers of the fig is known as caprification. It is effected through the agency of a small hymenopterous insect, *Blastophaga psenes* L. (*B. grossorum* Grav.), which according to Cotte and Reynier(8) parasitizes the gall flowers of the male or caprifig. Eisen(11), Vallese(43), Rixford(31), Condit(5), and most recently Grandi(15), (16), have given extensive accounts of caprification and its agent. A brief account of the process is given here. It consists in suspending caprifigs on the branches of the female tree at the time of the issuing of the insects. The female

blastophaga enters the female fig receptacle for the purpose of oviposition and in so doing carries pollen from the caprifig into the edible fig. Oviposition, however, is not effected, because, as has been already mentioned, the styles of the female flowers are much longer than the styles of the gall flowers where the blastophaga oviposits. The insect

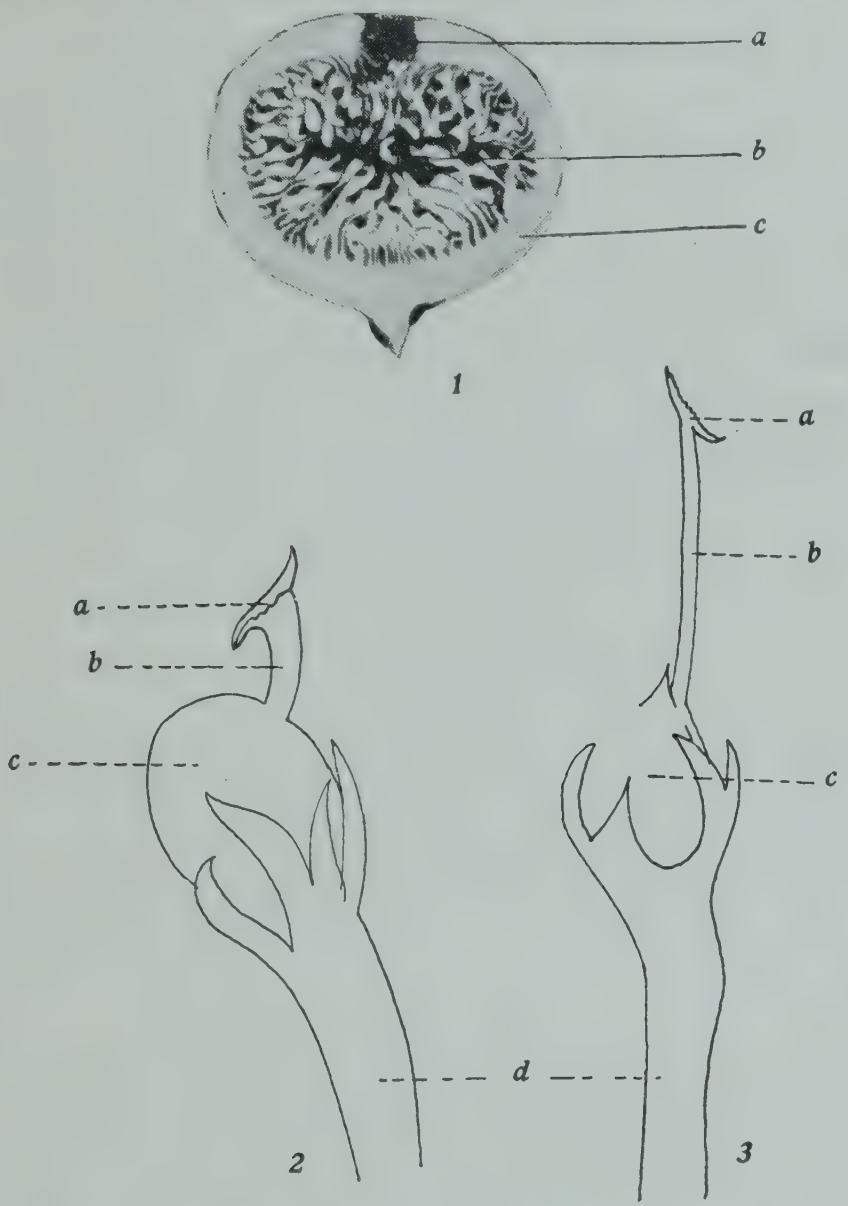


Fig. 1. Parts of the fig. (1) Longitudinal section of a Calimyrna fig. *a*, Osteolum or eye; *b*, pulp; *c*, meat. (2) Enlarged gall flower of caprifig. *a*, Stigma; *b*, style; *c*, gall; *d*, stalk. (3) Enlarged female flower of edible fig. *a*, Stigma; *b*, style; *c*, seed; *d*, stalk.

usually perishes in this vain attempt, but not without an effect, as the introduction of the pollen fertilizes the flowers of the varieties requiring such stimulation and causes the fruit to set and mature and form fertile seed. Without such a stimulation the fruit of the Calimyrna, San Pedro, and Stanford varieties drop when about one-

fourth grown, while those of the parthenocarpic varieties continue growth to maturity, but the seeds which they contain are not fertile (phenospermic).

In the gall flowers of the caprifig, the blastophaga inserts its ovipositor through the style and deposits a stalked egg in the ovule. The egg hatches and the larva lives inside the gall where it also pupates. The adult males issue first from their galls, fertilize the adult females while the latter are still in their galls and thus open the way for them to emerge later. The males, as a rule, never leave the figs. The females come out through the eye of the fig and while doing so rub against the staminate flowers which surround the orifice and shed their pollen at that time. The female insect may then carry the pollen into a fig of an edible variety or she may enter another caprifig, perhaps on the tree bearing the one from which she just emerged. In the latter case the pollen does not generally function as there are usually no receptive female flowers in the caprifigs. The term "caprification" is also applied to the setting of the male figs which results from the visit of the blastophaga.

DISEASES OF FIGS

It is only quite recently that the diseases of the fig have attracted the attention of investigators in California. The fig has been reputed to be particularly free from disease in most of the treatises on this fruit (Eisen[11], Roeding[32]). Foliage, twig, trunk, and root diseases have been reported from other states and from Europe, but, except a die-back of the twigs reported by Condit and Stevens(6) and Phillips(23), no disease of the tree has been known in California. Decline of the tree due to neglect, soil conditions, and nematodes has been discussed by Condit(4). Of the diseases of the fruit, a number occur in the South Atlantic and Gulf States and cause considerable damage. Edgerton(9) from Louisiana, Matz(21) from Florida, Stevens and Hall(41) from South Carolina, Potts(28) from Texas, and Gould(14) report a number of diseases of the fruit, the most important of which are an anthracnose caused by *Glomerella cingulata* (Stonem.) S. and v. S. (*Colletotrichum carica* Stev. and Hall) and a soft rot caused by *Rhizopus nigricans* Ehr. The first of these troubles has never been reported from California. *Rhizopus* has several times been isolated from rotted figs in the San Joaquin Valley.

Souring was the first disease of importance reported from California. It was attributed by Pierce(25) to an unidentified yeast but

no further work was done. Considerable time has been devoted to the study of this disease in connection with the present work; these findings will form the subject of another paper. A black smut due to *Sterigmatocystis* was reported by Hodgson(19) in 1918. An investigation of this disease was undertaken by Smith and Phillips(40) and a preliminary report was published in 1922. A more detailed account of this investigation was published by Phillips, Smith, and Smith(24) in 1925.

ENDOSEPSIS (INTERNAL ROT) OF THE FIG

Synonymy

The term 'souring' has been used indiscriminately in the past for almost every deterioration of the fruit of the fig. In careful observations, however, it is quite easily seen that symptoms vary considerably, and that there must be a number of agencies responsible for the large percentage of culls detracting from the profit of fig growing. The subject of this paper is a specific fruit-spoilage disease of the fig which has hitherto, except for a short note by the writer(2), remained undescribed and undifferentiated. At times a number of names have been used vaguely by growers to distinguish this trouble from what they consider typical souring, and the names 'pink rot,' 'brown rot,' 'soft rot,' 'stem-end' or 'eye-end rot' are frequently heard as referring to symptoms of disease observed in spoiled Calimyrna figs. All these names, although suggestive, seem either to be confusing or to apply only to certain phases of the disease. The names 'brown rot' and 'soft rot' are confusing because the first is applied to the well known disease of the stone fruits which does not affect the fig, and the second is applied to a rot of the fig in the Gulf States caused by *Rhizopus nigricans* Ehr. The names 'pink rot,' 'stem-end rot' and 'eye-end rot' refer to spots occasionally seen on the fruit, but as, in many cases, these spots are nothing but the external symptoms of a generalized disintegration of the meat and pulp of the fruit, they are not very descriptive. The name 'endosepsis (internal rot) is proposed here as more appropriate and less confusing.

Symptoms

The disease manifests itself internally as soon as the figs begin to ripen. In severe cases, even before the pulp sweetens and just as soon as the stalks and the sepals of the individual florets swell

and begin to color, brown streaks may be seen running down the flower stalks almost to the meat. As the fig ripens such streaks develop into spots, yellow-brown in color, and may involve a number of flowers. In most cases these colored spots are first found in the pulp near the eye of the fig, but any other part or parts of the pulp may develop this symptom according to the locus of infection. In very early stages, just as the fig begins to sweeten, these spots stand out very clearly against the bright-colored healthy pulp. Plate 1 shows six figs, five of which are at the same stage of maturity and alike in external appearance, firm, and bright green, just at the stage when figs are picked for canning or for fresh shipment. From their external appearance all five could be taken for sound ripe figs. Fruit *a* represents a healthy fig at this stage of maturity. The pulp should be bright amber to pink, with a small amount of sweet juice in the cavity. Fruits *b, c, d,* and *e* show the endosepsis symptoms at different stages of development. In fruit *b* the brown includes almost the entire mass of flowers in the vicinity of the eye, in the third fruit the browning includes three-quarters of the pulp, in the fourth all but a few flowers at the stem end are involved, and in the fifth the entire pulp is disintegrated. Such pulp is slightly watery and is easily pulled away from the meat. Until this last stage is reached, and even later, there is almost no external sign of this diseased condition of the pulp.

When the fig softens and begins to dry, a water-soaking of the skin appears in indefinite areas, mostly around the eye in a circular spot, or extending down the sides to the neck of the fig. This water-soaking gradually assumes a bright pink or purple color and the epidermis of the fig may easily be rubbed off on such water-soaked spots (pl. 2). The fig may dry in this condition and fruit *f* in plate 1 and the fruits in plate 3 represent such dried figs. These pink spots should not be confused with the pink spots produced by *Aspergillus niger* van Tiegh., the black-smut organism of the fig, as described by Phillips, Smith, and Smith (24). In the case of the internal rot the spots are not very wet and the margins are not shrunk and have not the tendency to be easily detached from the rest of the skin as in the black-smut spots. In the internal rot they appear as normal skin except in color. It is not always, however, that such colored spots appear on the decaying figs. Under favorable weather conditions the figs dry before they reach the stage of external symptoms, and undoubtedly even before the entire pulp is decayed. In many cases only a small water-soaked ring appears around the eye and a drop of liquid is exuded, varying in color from clear to caramel. This

exudate is never in sufficiently large quantities to drip and soil the foliage or solidify in long hanging drops, as is the case in souring. Many figs dry and pass for good fruit when the pulp is in reality full of the rot fungus. The interior is destroyed only in part and the flavor is not greatly affected. Such figs appear practically normal on the outside, although the inside is a little dry and "seedy" (see pls. 4 and 5) and the flavor slightly peculiar.

Finally, in certain orchards where the rate of drying has been slow on account of close planting, late irrigation, or climatic conditions, the disease becomes generalized in the fruit and the rotting of the pulp is so rapid as to separate the latter from the meat. The pulp slips easily from the meat and the fig appears sagging, wet, dripping, extremely soft and deformed. The pink spots may or may not develop on the sides, or end, depending on whether or not the parasite has actually invaded the meat.

The effects on the eating qualities of the fig produced by the disease vary considerably, depending largely on the bacterial flora present. Ordinarily, figs affected with endosepsis are lacking in odor and flavor rather than possessing disagreeable ones. The taste is rather flat, watery, lacking the proper sweetness and the characteristic fig flavor, with the seeds very prominent. There is no odor. In some cases, however, there is a very disgusting, putrid, somewhat bitter taste, very characteristic but impossible of description, and an odor which suggests that of spoiled tomatoes. There seem to be no ill effects from the eating of diseased figs.

Distribution and Economic Importance

Internal rot is distributed throughout the fig belt of California and wherever the Calimyrna is grown. No district seems to be entirely free from the disease and although it is confused with souring and the estimates of injury may be somewhat high, it is the opinion of the writer that this disease is very largely responsible for the size of the cull pile. Its severity depends largely on the climatic conditions, and the relation between these conditions and the disease will be discussed later on. However, it is due to this disease that successful growing of Calimyrna figs for the dry-fig market in certain regions and in certain years has become problematic. A large percentage of the dry product is either entirely unmarketable on account of the external pink discoloration, or the quality is lowered because of the internal deterioration of the pulp.

The loss given by the growers at different meetings in 1924 and 1925, and often probably greatly underestimated, varied from 20 per cent to 90 per cent of the dry crop. Specifically, the growers in one county in 1925 suffered from 20 to 50 per cent deductions for culls at a cannery on account of the internal rot. These shipments were made up of carefully selected fresh figs. An estimate of the percentage of dry figs showing *external* symptoms of the rot, as they were drying in the yard of one orchard, yielded a conservative figure of 70 per cent. The number of figs that had dropped to the ground from the several trees of another orchard was counted and the percentage of the figs showing external symptoms was found to average 30 per cent. At a meeting of fig growers held in 1924, the estimated percentage of loss from internal rot ranged from 33 to 71 per cent. If it is taken into consideration that the rot does not show actively until the fig has passed the canning stage, at which it must be firm, and that growers pick for canning only the best figs in the orchard, this percentage is alarmingly high. Many other counts and estimates were made at different times and the percentage of injury from this disease was always high.

Influence of Environment

It has previously been mentioned that climatic factors determine the severity of the disease. Such factors exercise no influence other than determining the rate of ripening and drying of the fig. This, in turn, determines whether the internal rot will become generalized in the pulp and also affects the appearance of external symptoms. Since, as will be shown later, infection takes place at a very early stage and the active fungous growths do not begin until the fig commences to ripen, the rate of ripening will determine the extent of damage to the fruit. That this is actually the case is shown by the percentages of disease observed in different localities and in different years. Sections with later and cooler seasons show always a larger percentage of damage than those which are warmer and dryer in late summer. This was especially well marked in 1925.

The amount of winter rainfall does not seem to be a factor, as in 1924 there was an alarming percentage of disease in all the sections, although the winter of this year was one of the dryest ever known in California. On the other hand, 1925 was a year of normal rainfall, but some sections showed a very small percentage of external symptoms of fig rot. The determining climatic influences are there-

fore the ones prevailing during July, August, and September. Quick ripening, rise in sugar content and concentration stops the growth of the fungus in the pulp and the fig dries without much loss in quality.

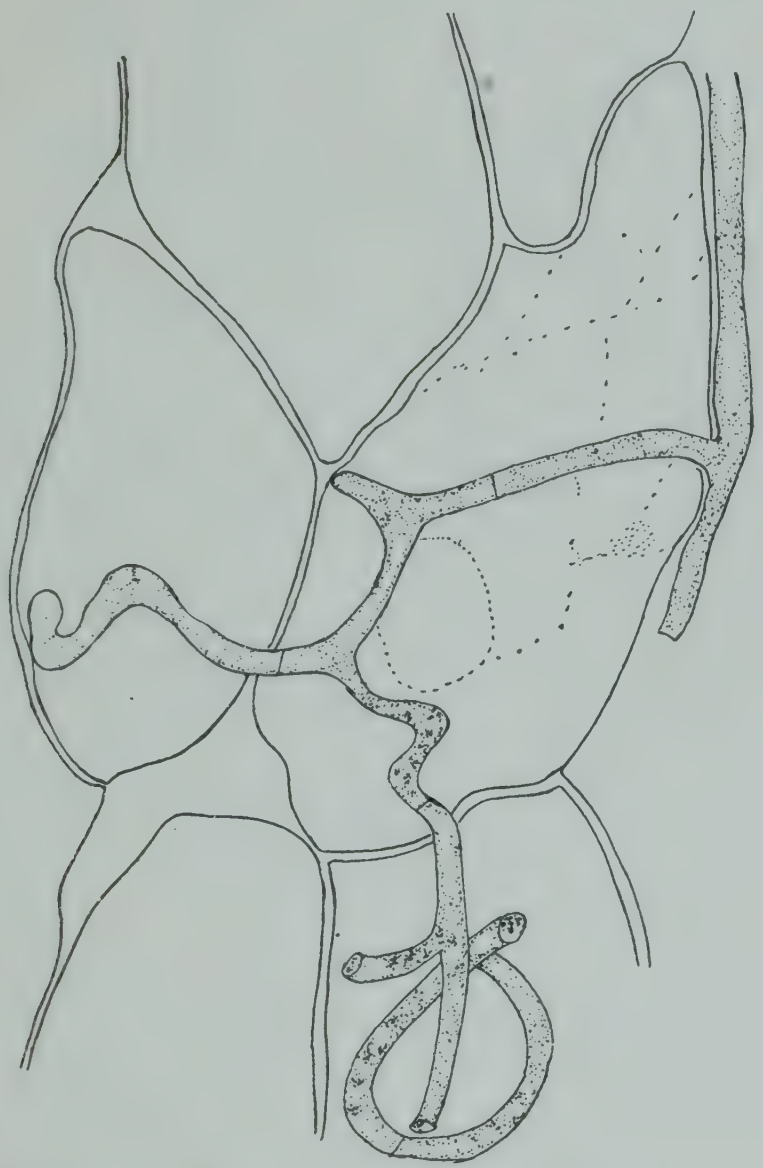


Fig. 2. Camera lucida drawing from stained preparations showing the penetration of the cells of the fig by the hyphae of *Fusarium moniliforme* Sheld. var. *fici* n. var. (× 1000.)

Etiology

Isolations.—The disease is caused by the fungus *Fusarium moniliforme* Sheld. var. *fici* n. var.

In a preliminary report(2) on this disease in the Calimyrna fig it has been stated that the diseased tissue is found permeated by a hyaline, septate mycelium of a fungus which can be very easily isolated and which grows luxuriantly on a variety of culture media.

The fungus does not usually produce aerial mycelium, either on the surface or in the cavity of the fig, but if a small portion of the rotting pulp is crushed under a cover glass and examined with the microscope it will be found permeated by a network of hyphae. The hyphae are more easily demonstrated by removing a small portion of water-soaked skin, dehydrating quickly in 85 per cent alcohol, staining in 2 per cent solution of Magdala red in 85 per cent alcohol, washing with absolute alcohol, clearing in xylol and examining under the low power of the microscope. The hyphae are colored red, while the host tissue is colorless or slightly pink (fig. 2).

If diseased figs are placed in a moist chamber they are soon covered by a woolly-white or pinkish, aerial growth. Figs, however, especially when ripe, form an ideal substratum for almost any saprophytic fungus; moist chamber studies, therefore, should not be relied upon for isolating the pathogen. Plate cultures from the pulp of a diseased fig which has not been exposed to the atmosphere always yield a typical growth or rather a group growth, which consists almost exclusively of the fungus mentioned and of a cream-colored and a bright red bacterium (pl. 6). This association has been found to be remarkably constant. It may be said also that both bacteria have been obtained from figs irrespective of disease symptoms, and in all stages of maturity, but not previous to caprification. These facts indicated a flora in the receptacle of the fig and an investigation was started to determine the flora of the edible fig in all the stages of development.

Phillips, Smith, and Smith(24) in 1925, in a study of fig smut, report that the pulp of Adriatic figs in early stages of maturity (stages 1, 2, and 3) before the fruit has been entered by insects, is entirely sterile. The classification of stages of maturity established as a basis for reference by Smith and Phillips in 1922(40) has been used in this work. Descriptions of the different types follow. For pictures of the different stages the reader is referred to Phillips, Smith, and Smith(24).

1. Fruit not quite full grown, still green and hard.
2. Full grown, eye scales beginning to loosen.
3. Eye fairly well opened, fruit still green and firm.
4. Slightly yielding to pressure, pulp succulent but still firm.
5. Fig ripe as for picking for fresh shipment. No shriveling, pulp opaque.
6. Skin slightly shriveled, pulp somewhat translucent.

7. Distinct shriveling, contents still red, not sticky.
8. Much shriveled and skin beginning to discolor; pulp mahogany color, slightly sticky.
9. Skin brown but flexible, pulp brown, translucent, sticky; stage of completed normal drying.

A repetition of the work of Smith and Phillips(40) regarding the flora of the Adriatic in stages 1 to 3 yielded identical results. The study was extended to figs of other varieties and the results indicated clearly that all parthenocarpic varieties, up to the time that they reach full growth and begin ripening as indicated by the loosening of the scales, coloring of the pulp and softening of the skin and pulp, are sterile. Varieties studied other than the Adriatic were the Black Mission (California Black), Brown Turkey, White Ischia, Kadota (Dottato), San Pedro White (first crop only), Cordelia, and miscellaneous other figs of unknown varieties. Both the first and the second crops were examined.

The method used was the standard poured-plate method with nutrient-dextrose agar, fig-infusion agar, or Czapek-dextrose-synthetic agar. The fig was washed with alcoholic $HgCl_2$ 1:1000, then split open with a sterile knife without touching the pulp, and a portion of the latter was scooped out and placed on the plate. A control plate was poured after every twenty-five inoculated plates. The Petri dishes were stacked and placed on a small platform in the center of a panful of water. A bell jar was then put over the stack of dishes with the mouth of the bell jar immersed in the pan of water. A U-tube was slipped under the wall of the bell jar so that one arm opened into the chamber and the other communicated with the outside, thus equalizing the air pressure. These precautions were taken to guard against excessive drying of the agar during the summer months in the interior valleys of California and to guard also against insects crawling in and contaminating the plates. The plates were thus kept reasonably free from contamination for a long time as indicated by the control plates remaining sterile. The plates were examined macroscopically ten days after pouring, and microscopically after another ten days. Figs were collected from different localities in the San Joaquin and Sacramento Valleys and the results of these investigations from 274 figs of different varieties are summarized in table 1.

Fifty-six Calimyrna first-crop figs not caprified were also found sterile. The second crop of the Calimyrna variety when not caprified was also sterile as shown by pouring plates both from figs that

dropped because of lack of caprification and also from figs in which caprification was prevented by bagging the twigs with manila paper hat bags a week before the blastophaga began issuing. Fifty-four figs were thus examined and were found to be sterile. When, however, the figs of any of the above mentioned varieties were caprified it was found that immediately after caprification their flora consisted of one or more of the three organisms mentioned in connection with isolations from diseased Calimyrnas.

TABLE 1

VARIETIES, LOCALITIES, AND NUMBERS OF PARTHENOCARPIC FIGS EXAMINED IN DETERMINING THE FLORA OF THE FIG RECEPTACLE

	Crop	Sacramento	Modesto	Fresno	Condition
Adriatic	First	4	10	53	All sterile
	Second	4	15	68	All sterile
Mission	First	3	.....	16	All sterile
	Second	.....	.....	18	All sterile
Kadota	First	.....	.....	20	All sterile
	Second	.....	.....	18	All sterile
San Pedro	First	.....	.....	18	All sterile
White Ishia	First	.....	.....	4	All sterile
Brown Turkey	First	.....	.....	4	All sterile
Cordelia	.....	10	.....	4	All sterile
Miscellaneous	.....	5	.....	.....	All sterile

Plate 7 shows the flora typically obtained from parthenocarpic varieties and from Calimyrna figs. The plates of the right hand series were poured from unripe, half-grown, fruits of Adriatic, Mission, and Kadota figs, respectively. The plates of the left hand series were poured from caprified Calimyrnas of the same stage of ripeness as that of the parthenocarpic figs used in the first series. It may be seen that there was no growth at all on the first series, while the fungus and the two bacterial organisms grew on the plates of the second series. Those discussed are typical of the many hundreds of plates poured during the course of this work. Plate 8 shows two Petri dishes, the upper poured with material from a caprified Adriatic fig, the lower with material from a non-caprified Adriatic. An abundance of growth is seen on the first plate, while the second one is sterile.

A great number of strains of the pathogen were isolated from diseased Calimyrna figs from several localities. The collection now includes over 250 strains which show more or less similar cultural characteristics. The majority of these strains are identical, the only difference being the origin or place of isolation. A great many of them exhibit variations of major or minor importance. The variations concern the habit of growth, size of spores, type of spores produced, color of mycelium, color of substratum, type of growth on different media, and lack of one or more of the following structures: Sporodochia, sclerotia, pionnotes, and catenulation of spores. Most of them, however, agree in a general way and evidently belong to the same group species, while three of them are quite different.

Eleven of the strains exhibiting the greatest differences were selected and studied intensively for over two years on a great number of media, including the ones recommended by Sherbakoff(36), the Fusarium Conference(47), Morris and Nutting(22), and others used for the first time in this study. Most of the differences were found constant. Certain strains, however, developed characteristic structures which were helpful in identification. The chief difficulty encountered in these studies was the stubbornly persistent failure of certain strains to produce the macroconidial type of spore characteristic of the genus *Fusarium* while their other characteristics clearly indicated such a relationship.

Morphology.—Strain 93 is considered as the type form of the new variety and has the following morphological characters. Microconidia in false heads or in chains formed on white to dark maroon purple[3] colored, aerial mycelium; ovoid-fusoid, 4.7–10.6 × 2.4μ.

Macroconidia delicate, slender, sickle-shaped, attenuate, subpedicellate, occur only in sporodochia on steamed corn meal and on woody stems, salmon-buff becoming wood brown with age. Sporodochia effuse, in columns, or in threads .5 to 1 mm. thick, twisting in loops or spirals 4 to 5 mm. long. Macroconidia mostly 3-septate (74 per cent) 19.9–49.4 × 2.3–7.4μ, 4-septate 39.9–49.3 × 3.52 to 4.1μ, 5-septate (rare) 43.5–51.7 × 3.5–4.7μ. Aerial mycelium dense fine or loose woolly, color varying from white to dark maroon purple, occasionally white with Payne's-grey-colored spots. Substratum light ochraceous buff becoming Vandyke red, Hessian brown or Perilla purple. Conidiophores simple on the sides of hyphae or di- and trichotomously branched to dendroid, alternately or oppositely arranged. Swollen cells (pseudochlamydospores) arranged in chains or singly are frequently found.

[3] The colors are after Robert Ridgway's color standards and nomenclature.

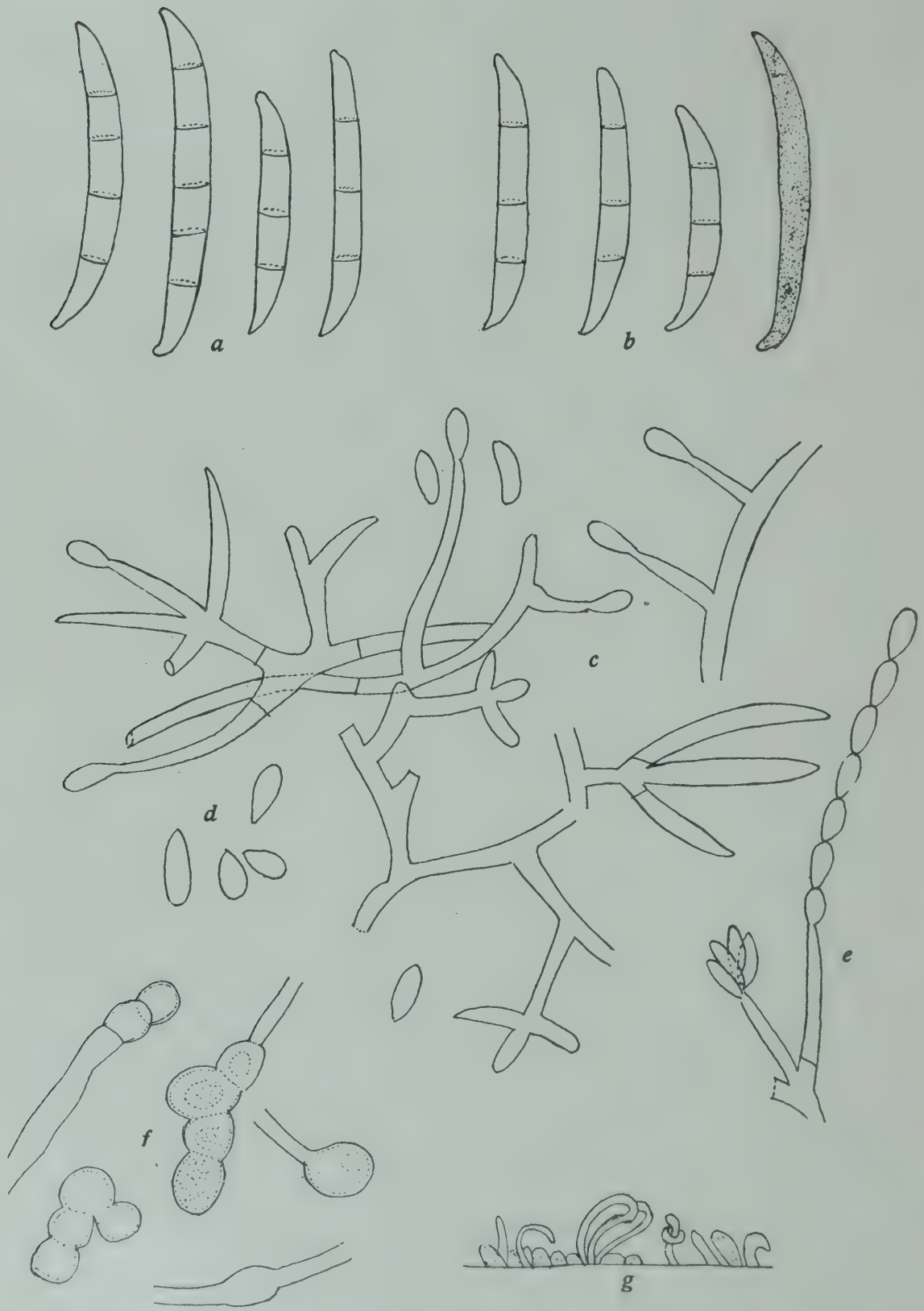


Fig. 3. *Fusarium moniliforme* var. *fici* n. var. *a*, Macroconidia from sporodochium on autoclaved cornmeal (× 782). *b*, Macroconidia from sporodochium on blackberry stem (× 782). *c*, Conidiophores and microconidia. *d*, Microconidia. *e*, Microconidia in chains and loose heads. *f*, Pseudochlamydospores (× 782). *g*, Sporodochia on autoclaved cornmeal.

Sclerotia are rarely produced on steamed rice and on hard oat agar. This characteristic is more pronounced in some of the strains, while in others it is entirely absent. The sclerotia are very small (1 mm. in diameter) and greenish black in color on rice, but larger and lighter in color on oat meal.

What is apparently the perfect stage of the fungus has been observed only once, on a plate from a Milco variety mamme (capri) fig collected from a tree at Ripon, California. Nothing unusual was noticed at the first observation of the plate, ten days after pouring. The flora on the plate was typical of hundreds of plates. But on a reëxamination of the plate sixty days after pouring, it was found that a semicircular row of black bodies had formed at the point of contact between two colonies of *F. moniliforme fici* originating from fragments of gall flowers. On a preliminary examination of this plate the following observation was recorded: "White fungus at a corner from flowers, the rest sterile." This refers to colony *b* (plate 9), which started from small fig fragments. Colony *a* is either a contamination or a later development from the large fig fragments in the plate. The black bodies referred to above proved to be perithecia. Scattered perithecia were found on the intermixed hyphae. The perithecia are black, osteolate, gregareous or solitary, containing ascospores which are greenish, thick-walled, usually 1-septate, measuring $10.15 \times 5.1\mu$ ($12.3–8.2\mu$). Germination of the ascospores is rather low. Isolation of the two parent colonies was made but repeated attempts on different media to induce the production of perithecia failed.

Nomenclature.—In a preliminary report(2) on this disease in the Calimyrna fig the organism constantly isolated from diseased figs was tentatively identified as *Oospora verticillioides* Sacc. and described as "having a hyaline, frequently branching septate mycelium . . . fruiting abundantly, producing catenulate, short or long, tapering or slightly curved, unicellular conidia on single or branched conidiophores borne on the sides of the hyphae." In a recent paper by Wineland(45) an ascigerous stage was ascribed to *F. moniliforme* Sheldon and its synonymy critically analyzed. There is considerable evidence given to indicate that *Oospora verticillioides* Sacc. is a synonym of *F. moniliforme* Sheldon, although there is no direct proof of such a relation.

Chen(3), in studying the internal fungous parasites of agricultural seeds, has isolated from seed corn a fungus which according to his description closely resembles the fungus isolated from figs. Chen

did not find macroconidia of *Fusarium* in his cultures, therefore the fungus was identified by him as *Oospora verticillioides*. He states that "should *Fusarium* spores develop, the parasite would have to be referred to *Fusarium moniliforme* Sheldon." Fusarium spores were found in these cultures later by Wineland(45). Both *Oospora verticillioides* and *Fusarium moniliforme* have repeatedly been reported as parasites or saprophytes on corn, the first from Europe by Deckenbach in Bessarabia and by Cuboni, Tiraboschi, and others in Italy; and the second in America by Sheldon, Valleau, and many others. The relation of these fungi and references to the literature were recently given in detail by Wineland(45). The experience of investigators working with *Fusarium moniliforme* tends to show that macroconidia are not always produced, and this has been my experience. In the studies on the distribution of the fig disease throughout California, I poured more than four thousand plates from caprifigs and parthenocarpic and caprified figs, and had occasion to study several thousands of colonies of the fungus causing the rot. Macroconidia were never seen in any significant number in aerial mycelium, nor were sporodochia, pionnotes, or related structures ever observed on these plates. The media used in this study were mostly nutrient-dextrose agar, Czapek-dextrose-synthetic agar, fig-infusion agar, and potato-decoction agar. Macroconidia were, however, obtained in a special study and only from certain of the strains of the fungus. Macroconidia were also observed on the surface of Czapek-dextrose-agar and fig-infusion-agar dilution plates soon after the germination of the microconidium. These macroconidia, however, were soon overrun by aerial mycelium and after this only microconidia were produced on these media. Strains producing no macroconidia would have to be referred to *Oospora verticillioides*, at least tentatively, until macroconidia are obtained. The production of macroconidia in culture would place such fungi in the genus *Fusarium*.

In order to be sure that no mistakes were made in the identification of the fungus, a strain of *Fusarium moniliforme* Sheldon was requested from Miss G. O. Wineland of the U. S. Department of Agriculture, who has recently studied(45) the species exhaustively, and another from Dr. E. A. Bessey of the Michigan Agricultural College. The strain received from Dr. Bessey bears the number 299 and was originally received in 1924 from Dr. C. L. Shear of the U. S. Department of Agriculture under number 884. This culture was isolated from banana in Honduras and was sent to Dr. Shear by Dr. O. A. Reinking. Miss Wineland's culture bears the number 16 and was sent

as the most typical of the species from the strains studied by her. Diseased corn kernels were received from J. B. S. Norton of the Maryland Agricultural Experiment Station and from them was isolated a fungus producing catenulate *microconidia* in sporodochia. *Fusarium moniliforme* was also isolated from diseased ears of corn sent from Stockton, California, to Prof. W. T. Horne of the plant pathology laboratory of the University of California. All these strains were grown in parallel cultures with several strains of the fungus from figs and carefully compared on a great variety of media.

In some respects the fig fungus agrees fairly well with the descriptions given for *F. moniliforme* Sheld. by Sheldon(35), Wollenweber and Reinking(48), and Wineland(45). It differs, however, in the minimum measurements of all forms of spores, the fig fungus having shorter micro- and macroconidia. The fig fungus shows more aerial, more highly raised, and looser mycelium on some media. The aerial mycelium is more highly colored on some media and it has never been seen to be Isabella color or even any related color. The colors are white, pink, maroon, or reddish purple. Sporodochia are not usually produced on ordinary media, and when produced on steamed corn meal and elder stems they are formed in columns, or are filiform, suggesting possible catenulation. Macroconidia are not found in aerial mycelium. Sclerotia are not produced by the majority of the strains. Swollen cells in chains or singly are found in the mycelium; no pseudo or true pionnotes forms were ever observed. This form therefore is considered as a new variety, being named *Fusarium moniliforme* Sheldon var. *fici* n. var.

Cultural Characters.—As mentioned previously, many of the strains were grown on a variety of media in an effort to induce the production of macroconidia in culture. Strain 93 described above varied considerably on the several media employed and its characteristics on certain standard media will be described and measurements of the macroconidia given.

The media used were Coons's, Czapek's dextrose, Czapek's sucrose, and Sideris' synthetic agars; hard oat, soft oat, fig, lima bean, prune, potato, and corn-meal decoction agars; blackberry, tomato, *Melilotus,* fig, and elder stems; potato, fig, carrot, and coconut plugs; corn meal, rice, and oat meal in 150 c.c. Erlenmeyer flasks, and in tubes; crackers in deep Petri dishes and hard potato decoction agar with the addition of 5 per cent dextrose in Petri dishes to determine color production and rate of growth.

The most valuable of these media were: Coons's synthetic, hard oat, and fig agars in slants, blackberry and elder (*Sambucus nigra*) stems, potato plugs, rice in test tubes, oat and corn meal in 150 c.c. Erlenmeyer flasks, and hard potato plus 5 per cent dextrose agar in Petri dishes. These media were made according to directions given by Sherbakoff(36), Coons(7), and Sideris(37). Fig agar was made by boiling 100 gms. of dried figs for 30 minutes, straining, filtering, adding 2 per cent agar and sterilizing at 15 lbs. pressure for 20 minutes. Cornmeal and oatmeal flasks were prepared by adding one part of the meal by volume to three parts of water and steam sterilizing for one hour on three successive days.

Coons's agar: The aerial growth was scanty, powdery, white, dotted with salmon-colored sporodochia. The color on the substratum was Bishop's purple, non-diffusing. The mycelium and the spores were very granular and the septation in the macroconidia indistinct. 1-septate and 0-septate macroconidia were found; 0-septate macroconidia measured $8.2 \times 3.1\mu$; 1-septate, 16.4μ; 3-septate $28.7 \times 4.1\mu$. Swellings occurred in the hyphae.

Hard-oat agar: The aerial growth was hydrangea-pink, downy, pinkish vinaceous, abundant, with sporodochia in groups. The medium was dark mineral red. Swellings occurred in the hyphae.

Fig. agar: The abundant powdery growth ranged from pinkish vinaceous to dark Corinthian purple. The medium was Hay's maroon. No sporodochia were found.

Blackberry stem: The fine white growth was pale vinaceous fawn with dull Indian purple spots and the large scattered sporodochia were salmon buff. Macroconidia in sporodochia were mostly 3-septate, $28.2–37.6 \times 3.4–4.7\mu$ (average $33.0 \times 4.2\mu$); microconidia measured 7.0 to 8.8μ long. Sporodochia are not produced when there is much water in the tube. Certain strains produced sporodochia containing microconidia exclusively.

Elder stems (*Sambucus nigra*): Abundant aerial white growth developed. Many sporodochia were found, in color between light and pale ochraceous buff, gregarious and solitary, effuse. Two hundred macroconidia were studied regarding septation, using the method suggested by McWhorter(20). The percentages were as follows: 0-septate, .5 per cent; 1-septate, 10 per cent; 2-septate, 5 per cent; 3-septate, 74 per cent; 4-septate, 10.5 per cent; no 5- or 6-septate spores found. Microconidia measured from 4.7 to 10.6μ in length (average 7.8μ); 3-septate macroconidia measured 28.2 to 33.7μ in length (average 28.2μ).

Potato plug: Abundant matted white growth developed with the substratum blue in spots. Macroconidia were produced occasionally, but these were seldom in columns. Swellings occurred in the hyphae.

Steamed rice: After 30 days, there was abundant aerial white growth. Interstices were pinkish vinaceous, kernels light Corinthian red. In sixty days the interstices turned Corinthian pink, the kernels Corinthian red and Indian red. When a smaller amount of water (1:1) was added to the rice the colors were purple drab to pallid vinaceous.

Cornmeal flask: This medium was found very valuable for the production of normal macroconidia. The aerial growth upon it was abundant and white, slightly tinged with salmon. Sporodochia, salmon-colored and in tendrils or columns, developed in great abundance after some time (50 days). The macroconidia measured as follows:

1-septate, 12.9–16.4 × 2.3μ (average 14.2μ).
3-septate, 20–40 × 2.4–4.1μ (average 30.5 × 3.4μ).
4-septate, 40–50 × 3.5–4.1μ (average 46.5 × 4.0μ).
5-septate, 43.4–51.7 × 3.5–4.7μ (average 46.8 × 3.7μ).

Oatmeal flask: Abundant growth was made, shell pink, slightly vinaceous in color. An abundance of sporodochia developed as in cornmeal, salmon or greenish-blue in color. Pseudochlamydospores occurred in abundance on the mycelium.

5 per cent dextrose hard potato agar in Petri dishes: In the dark, aerial mycelium was very woolly and raised, abundant, white to pale grayish vinaceous; on the medium dark Perilla purple to maroon. Growth from center of Petri dish to the edge (5 cm.) was made in six days. Spores occurred in chains but mostly in loose balls on the end of the much-branched conidiophores.

The organism was grown on these media repeatedly with identical results.

It is mentioned by Wineland (45) and several other investigators that strains of *Fusarium moniliforme* when grown in culture for a long time or when only mycelium or microconidia are used in the transfers, lose their power of producing macroconidia as well as their chromogenic properties. The strain just described of the organism from the fig has been under culture for almost three years and transfers of it have been allowed to dry for over a year at high temperatures. When transferred to suitable media they did not seem to differ in the least from the original descriptions. However, the same cannot be said of other strains of this fungus.

In the course of this work it was suspected that light and the moisture content of the medium exercised an effect on the color formation and the production of sporodochia. To test this point 8 gm. portions of cornmeal were put in each of seven 150 c.c. Erlenmeyer flasks and water was added in increments of 4 c.c., from 8 c.c. of water up to 32 c.c. The series was prepared in duplicate. The flasks were autoclaved at 15 pounds pressure for 20 minutes, cooled in the refrigerator and immediately inoculated with .5 c.c. of a heavy suspension of both macro- and microconidia. One series was kept constantly in the dark at room temperature while the other was exposed to a rather weak diffused daylight near a window of northern exposure shaded by large eucalyptus trees. After 20 days the growth was profuse on all the members of the series, but sporodochia developed in great abundance only on the members of the series exposed to the light and in inverse proportion to the amount of water initially added. A suspicion of sporodochia only showed in the members of the series not exposed to light that contained 8, 12, and 16 c.c. of water respectively. On the other hand, the color developed on the medium was very much more intense and bright in the series kept in the dark and especially so in the members of the series that contained 24, 28, and 32 c.c. of water, respectively. These results are summarized in table 2.

TABLE 2

EFFECT OF LIGHT AND WATER CONTENT ON COLOR AND SPORODOCHIA PRODUCTION OF F. MONILIFORME VAR. FICI GROWN ON CORNMEAL

Water (cc.)	Diffused light		Dark	
	Sporodochia	Color of substratum	Sporodochia	Color of substratum
8	++++	Flesh pink	−	Shell pink
12	++++	Flesh pink	±	Light Corinthian red
16	+++	Light Corinthian red	±	Buff pink
20	++	Russet vinaceous	−	Buff pink
24	++	Onion skin pink	±	Deep Corinthian red
28	+	Onion skin pink	−	Deep Corinthian red
32	+	Onion skin pink	−	Deep Corinthian red

This is in accordance with the findings of Sherbakoff (36) regarding the intensity of color produced by *Fusarium* grown in darkness. Coons (7) has found that diffused light is necessary for the pycnidia formation of *Plenodomus fuscomaculans*.

The effect of the hydrogen-ion concentration on the development of pigment in *Fusarium* has been recently discussed by Sideris(38) with a review of the literature on the subject. He found that *Fusarium moniliforme* Sheld. produced a range of diffusible colors from flesh to colorless when grown in liquid media with adjusted H-ion concentration ranging in pH from 3 to 7.5. This reaction was maintained constant by additions of acid or alkali throughout the growth period. When the reaction was not kept constant the colors ranged from hydrangea pink to lilac, indicating that in the alkaline members of the series where no color was produced when the reaction was maintained constant, pigment developed through the shifting of the pH by the metabolic products of the organisms.

Fusarium moniliforme var. *fici* (strain 93) was grown in Petri dishes on a series of solid media ranging in pH from 4 to 8. The media were prepared according to the directions of Sideris(38). Ten c.c. portions were poured and the dishes were kept in the dark. The rate of growth and the pigment development was frequently noted. No difference in the rate of growth was observed. The organism grew equally well and fast on plates of pH 4, 5, 6, 7, and 8. The pigments, however, differed on the different members of the series and also from those given by Sideris(38). Table 3 presents the colors diffused through the agar.

TABLE 3

EFFECT OF HYDROGEN-ION CONCENTRATION ON PIGMENT FORMATION IN FUSARIUM MONILIFORME VAR. FICI

pH Initial	Color after 6 days diameter of disc 30 mm.	Color after 14 days diameter of disc 45 mm.	Color after 33 days diameter of disc 50 mm.
4	Maroon to Van Dyke red fading to hydrangea pink	Victoria lake to Van Dyke red	Burnt lake
5	Dark Indian red to vinaceous rufous to salmon	As in 4 but brighter	Burnt lake
6	As in 4 for one-half of disc; as in 5 for remainder	Perilla purple to colorless	Burnt lake
7	Black to purplish-lilac to colorless	Neutral red to colorless	Dark Corinthian purple
8	White except on center which is dark blue slate	Deep livid purple to colorless	Indian purple

Strain 16 received from Miss Wineland was also grown in a similar series. The colors corresponded to those given by Sideris, viz., hydrangea pink to salmon. In the fig fungus the pigment is of the brighter maroon on the acid side, grading at first to dark reddish purple on the alkaline side; the colors of the alkaline side approach those of the acid side when the reaction is increased in acidity because of the action of the fungus.

Associated Organisms.—Two bacterial organisms, a red chromogenic and a colorless form, were found as constant associates of *Fusarium moniliforme* var. *fici* in diseased tissues of the fig and also alone in caprified figs, both ripe and green. The red chromogen was isolated the greater number of times from figs in the San Joaquin Valley. The colorless organism was more often found in figs from the Sacramento Valley, but both organisms were many times obtained from the same fig throughout California.

Morphologically the two organisms appear identical. They are both short rods $1 \times .8\mu$, arranged singly, asporogenous, motile, staining easily with ordinary stains, Gram negative. Culturally they are also identical except for chromogenesis. On agar slant at 30° C. they make an abundant, filiform, raised, glistening, opaque, viscid growth and have a decided odor which is similar in both and rather putrid. They liquefy gelatin rapidly. There is no surface growth on nutrient broth; the clouding is slight, the odor decided and the sediment scant. The liquid is colored red by the chromogenic form. They both possess a slight diastatic action, and produce an alkaline reaction on milk in two days with a rennet curd and peptonization. There is no reduction of litmus. They ferment dextrose and sucrose with the production of gas and acid (pH 4.5) but do not ferment lactose. Indol is not formed and nitrates are strongly reduced to nitrites.

The white strain answers the description of *Achromobacter nitrificans* (Burri-Stützer) Bergey(1). The chromogenic strain could not be assigned to any one of the species of Serratia described by Bergey(1). It differs in a number of ways from the type species *Serratia marcescens* Bizio (*Bacillus prodigiosus* Flugge); namely, in the alkaline reaction with rennet curd and peptonization in litmus milk, in the thick, raised, undulated margin of the colonies on agar, and in the fact that it has never been observed to form filaments or chains.

Both the red and the white organisms were found rather polymorphic on agar plates, especially when developing from fig tissue.

Plate 10 illustrates some of the differences. In dilution plates the red organism produces round, raised, glistening colonies with smooth margins. Such colonies are seen at the lower left of Petri dish 1109 in plate 10. It soon, however, begins spreading in fan shape, producing a colorless edge. This is more characteristic when the organism spreads from fig tissue placed in the center of the Petri dish. It then spreads in slender, faintly pink streams which unite, producing an undulate, dark red edge (Petri dish 1084, plate 10).

The appearance in nature and in the same habitat, of chromogenic and colorless forms, otherwise physiologically and morphologically identical, appeared to the writer to be of interest in the light of modifications and variations experimentally produced by selection or otherwise in *Serratia marcescens* Bizio (*B. prodigiosus* Flugge). Scheurlen(34) obtained colorless strains of *B. prodigiosus* by growing the organism on potato and selecting always from the least colored portion of the growth for subsequent transplanting. Wolff(46) also obtained long white forms of *B. prodigiosus* by careful selection and repeated transfers. Rettger and Sherrick(30) developed through selection white and red strains of *B. prodigiosus* from a culture which was weak in chromogenic properties. The chromogen from the fig has never shown weakness in chromogenesis, although it has been grown on acid media and has been kept without transferring for a long time; similarly, pigmentation has never been observed in the white strain.

Porter(27) found that *Fusarium lini* was inhibited in its growth by a bacterium. The fig fungus does not seem to be affected by the proximity of growth of either the white or the red bacterium described above, as can be seen from plate 6.

Pathogenicity.—Experiments to determine the pathogenicity of the organisms isolated have met with many difficulties. Inoculations could not be made on Calimyrna figs, since they may have already become infected in the process of pollination (caprification) which is essential to the development of the fruit of this variety. Fortunately it was observed that when Adriatics contract the disease through chance caprification they exhibit the same symptoms as Calimyrnas. Normally, and unlike the Calimyrnas in this respect, they develop fruit without caprification and on repeated examination were found to be sterile up to the time of ripening. Through the kindness of Mr. Taylor of the Pasa Rica ranch near Fresno, a row of large Adriatic trees was put at the disposal of the writer for inoculation purposes.

Keeping inoculated figs in moist chambers was found to be impractical because as soon as the ripe fig is put into a humid atmosphere it is invaded by a variety of saprophytes. The method of removing the glass lids and covering with cheesecloth was tried. This was found unsatisfactory because the figs dried too fast to show maximum development of the rot. It was finally found that by inoculating the figs at stages 4 or 5 of development, while still attached to the twig, and then bagging the entire twig with a three-pound-size manila grocery bag, satisfactory results could be obtained. The figs were thus inoculated while still sterile, since the eye is scarcely opened at stage 4, and were subsequently protected by the paper bag from wind and insect-borne infections as well as from excessive and rapid drying. No inoculated figs could be lost, since when they ripened and dropped they remained in the bags.

The entire collection of *Fusarium,* including approximately 250 isolations and the collection of other miscellaneous organisms including a variety of green fungi (*Cladosporium* sp., *Alternaria* sp., *Helminthosporium* sp., *Hormodendrum* sp.), and other moulds which appear occasionally on the plates, as well as the red and white bacteria described above, were inoculated, in duplicate, into figs. The inoculations were made by introducing mycelium and spores through the eye of the fig by means of a sterile platinum needle. The eye of the fig had been previously sterilized by swabbing it with a piece of cotton wetted in an alcoholic solution of $HgCl_2$.

The results have been very striking and conclusive. The typical symptoms of endosepsis, including the disintegrated pulp, and water-soaked purple or pink skin spots, the eye and stem-end rot, the "slip-skin" condition and the yellow gum at the eye were produced by the inoculations of strains of the *Fusarium moniliforme fici.* The green fungi and miscellaneous organisms were not found to be parasitic on the fig. Their growth when inoculated did not progress beyond the point of inoculation. They occasionally fill the cavity with a mass of superficial mycelium, utilizing the free sugar in the juice of the ripe fig; *F. moniliforme fici,* on the other hand, penetrates and disintegrates the pulp and the meat and permeates the affected tissues in every direction. Plates were poured from all the figs inoculated and in every case the organisms were regained in pure culture.

Examination of these plates showed that no changes had been effected in the organisms by passing them through the host; i.e., strains weak in chromogenesis or fruiting did not regain such lost properties or acquire others. A greater amount of aerial mycelium

was found in inoculated figs than is usually found in natural infections, but this is probably due to the large amount of inoculum introduced into the cavity.

Twenty sterile Adriatic figs were inoculated with the red and the white bacterial organisms, both in moist chambers and on the tree. The organisms were readily regained from the inoculated figs twenty-five days after inoculation but no change was observed in the fig. This is in accordance with what was observed in isolation experiments. Normal-appearing figs showing absolutely no deterioration yielded either the red or the white organism when plated. Finally, numerous inoculations were made by using one of the strains of the fungus together with either the red or the white bacterium. The results have not been very conclusive but suggest that the bacteria help in the disintegration of the pulp and contribute to the odor but cannot initiate the rot.

Transmission

The Flora of Caprifigs.—The difference in the flora of caprified and non-caprified edible figs suggested a study of the flora of caprifigs. The methods used in this work were the standard pathological ones. Caprifigs in all stages of development, before and after caprification (pl. 11) were collected from many parts of California. Usually not less than five figs were taken from one tree and not less than twenty-five from one orchard at one time. The methods used were those previously described. Duplicate plates from the same fig, using the same or a different medium, were often poured, but finally, as the flora was identical in both plates, and examination of a greater number of figs was desirable, a single plate was poured from each fig. However, care was taken to sample the pulp in such a way as to obtain a correct picture of its flora. A blank plate was poured as a check for every twenty-five plates.

The flora of caprified male figs was surprisingly uniform. At times a variety of saprophytes, usually *Alternaria* and other green fungi, was found, but not in such a way as to indicate a definite connection with the disease occurring in the edible figs. A variety of bacteria was also found in the course of the work, but two organisms, the red and the cream-colored one, were almost constantly obtained from the pulp of caprifigs. These two organisms were the same as those constantly isolated from Calimyrna figs. The fungus associated with the internal rot of the Calimyrna was also frequently isolated from caprifigs. Table 4 presents in a tabular form the flora of these caprifigs, as it was obtained from the different sections of California.

TABLE 4

THE FLORA OF CAPRIFIED MALE FIGS FROM COMMERCIAL ORCHARDS IN CALIFORNIA DURING THE YEARS 1922, 1923, 1924, AND 1925

	Number of figs examined	Per cent *F. moniliforme* var. *fici*	Per cent red bacterium	Per cent white bacterium	Per cent green fungi	Per cent Torula yeast	Per cent sterile figs
Mamme 1922 and 1923:							
Sacramento Section	52	30.7	13.4	61.5	.....	11.5	19.2
Modesto	45	35.5	40.0	35.6	2.22	2.22	20.0
Merced	22	54.6	63.6	59.0	4.55	4.55	9.08
Fresno	29	31.0	44.8	34.5	3.45	3.45	17.2
Reedley	49	32.6	61.2	28.6	18.4	18.18	2.04
Tulare	24	41.7	70.8	62.4	4.17	.....	16.6
	221						
Profichi 1923:							
Modesto	63	46.0	68.3	44.4	12.7	.....	4.76
Merced	45	51.1	82.1	40.0	6.66	.....	4.45
Fresno	115	40.9	65.2	18.3	10.4	.....	13.9
Reedley	46	52.2	78.2	21.7	4.35	.....	.....
Tulare	54	46.3	92.5	38.9	3.70	.....	7.41
	323						
Profichi 1924:							
Modesto	16	50.0	43.8	68.7	25.0	12.5	6.25
Fresno	43	69.8	67.4	67.4	.....	.....	.....
Reedley	47	51.1	80.8	51.0	4.25	8.50	....
Tulare	44	25.0	43.2	51.0	.....	.....	6.81
Southern California	70	17.1	.....	.....	.....	11.40	10.00
Mammoni 1924:							
Sacramento	79	55.6	.....	.....	.....	.....	.....
Modesto	12	58.3	41.7	.....	.....	.....	.....
Fresno	5	100.0	.....	.....	.....	.....	.....
Reedley	7	28.6	.....	100.0	.....	.....	.....
Southern California	5	60.0	.....	.....	.....	.....	.....
Mamme 1924:							
Sacramento	500	59.4	.....	.....	.....	.....	.....
Modesto	64	45.3	20.3	23.4	.....	.....	.....
Total Mamme	785						
Total Profichi	543						
Total Mammoni	108						
	1,436						

The State is divided for convenience into seven sections: the Sacramento Valley section; the Modesto section, comprising the Modesto, Ceres, and Turlock fig orchards; the Merced section, comprising the Merced, Planada, and Le Grand fig orchards; the Fresno section, comprising the Fresno, Figarden, and Clovis fig orchards; the Reedley-Dinuba section; the Tulare section, comprising the Orosi, Farmersville, Lindsay, and Strathmore orchards; and the southern California section, comprising the orchards south of Tulare and including the Coachella and Imperial valleys. During the last two years, after the relation of the fungus to endosepsis had been definitely established, media favoring the growth of fungi were used rather than neutral media permitting a great bacterial development. For this reason the percentages relating to the presence of the bacteria usually found in caprifigs are low or lacking. The red organism does not develop color on acid media; it forms pin-point colonies which grow very slowly, and their differentiation and identification is difficult.

This table shows that the forms of *Fusarium moniliforme* var. *fici* (*Oospora verticillioides*) which are always present in cases of endosepsis, together with the two bacterial forms, constitute what might be termed the normal flora of the male fig in its entire succession of crops, except in non-insectiferous caprifigs. The percentages of occurrence vary considerably but this is to be attributed to the small number of figs examined from one place at one time. The total number of figs examined is large enough to preclude chances of error. The fungus and the bacteria have been obtained from caprifigs and edible caprified figs at all stages of development, but only after the entrance of the blastophaga.

In certain varieties of caprifigs, for example, Roeding No. 1, Roeding No. 3, and Milco, some of the fruit of the profichi crop remains on the tree and develops normally, producing pollen, although it does not contain insects. Such figs are commonly known as blanks and can easily be distinguished from the insectiferous figs by their smaller size and lighter color. Blank figs in different stages of development were used in pouring a large number of plates but they invariably proved to be either sterile or to possess a saprophytic flora distinct from the one occurring in caprified figs. Caprifigs as a rule do not soften or acquire sugar as do the edible figs, except in a few varieties like the Milco and certain seedlings and only in the mammoni crop. On account of this fact the symptoms of the disease are not clearly defined as in the edible fig. The profichi crop, which is the most abundant of the three, is usually picked before ripening and

hung in the edible fig trees in baskets to facilitate caprification. These profichi figs gradually become hard and dry. If such dried profichi are split open, they are found filled with the aerial mycelium of the fungus previously described which has developed in the cavity after the blastophagas issued and before the figs became bone dry. Occasionally profichi were found exhibiting a wet and mushy internal rot and in many cases no insects issued from such figs. The external symptoms of the disease, i.e., bright pink or purple spots around the eye and on the sides, have been observed on fleshy mammoni figs of the Milco and Maslin varieties.

The Blastophaga as Carrier.—The fact that the fungus was obtained from both Calimyrna and caprifigs and not from any of the non-caprified figs pointed to the only connecting link between the two, viz., the blastophaga. In order to study this point, caprifigs from an orchard showing a high percentage of endosepsis were taken to the laboratory just before the blastophagas issued. The eye and the surrounding skin of each were carefully sterilized in 1:1000 $HgCl_2$ in 50 per cent alcohol and a sterile homeopathic vial was attached to the eye of each fig by means of melted paraffine (pl. 9). The figs were placed in a test-tube rack in diffused light until the blastophagas issued. When about a dozen insects had collected in each vial, the vials were detached and the insects removed one by one by means of sterilized forceps and placed on a poured Czapek-dextrose-synthetic-agar plate. Care was taken to immerse the insect in the agar so that movement was stopped. The fig from which the insects had issued was also used in pouring a plate so that the presence of the fungus in the cavity of the fig could be ascertained. Media permitting the development of bacteria were also used and the experiment repeated a great number of times using caprifigs from different sources and crops. These experiments proved plainly that the blastophagas actually carry the organisms discussed from the caprifigs into the edible figs at caprification time. The flora of each fig as determined by pouring a plate was also obtained from the blastophaga, caught as previously described. All combinations of the three organisms were found to be carried by the insects. Plates 12, 13, and 14 show some of the many plates poured in these experiments. The lower Petri dish in plate 12 shows the development of a colony of fungus from each insect placed on the agar plate. The same results were obtained when individual gall flowers picked one by one from the cavity of the caprifig, were placed on another agar plate as shown in plate 12 (upper). The Petri dishes in plate 13 illustrate the development of

both the red and the white bacteria as well as of the white fungus when insects or gall flowers are placed on poured agar plates. However, not all the blastophagas, even from a fig that showed the white fungus, were found to carry that organism, but almost all of the blastophagas plated carried at least one of the two bacterial organisms.

Method of Transmission.—Insects have for a long time been known as carriers of plant diseases. Rand and Pierce(29) have given an extensive review of the whole subject of insect transmission in plant and animal diseases. They found that insects transmit pathogens in three ways: (1) mechanically, by picking up the spores on the exterior of their bodies and accidentally sowing them on the surface or inoculating them into punctures; (2) by making avenues of infection through wounds; (3) by transmitting them internally, either mechanically or biologically.

In our investigations it was found that the pathogen and the organisms associated with it are carried by *Blastophaga psenes* L. Extensive studies were made in order to ascertain the method of transmission, which might in this case be "external-mechanical," "internal-mechanical" or "internal-biological." The life of the adult blastophaga is very short. Grandi(15) has kept them alive in captivity for 4–5 days and Vallese(43) for 8 days. The female, after laying its eggs, usually dies in the cavity of the fig where the eggs were laid. The male usually does not come out of the fig where he lives. Neither the male nor the female adult insects have been observed feeding on any of the parts of the fig. Grandi(15) has found that they possess well-formed mouthparts which are used in opening their way out of the gall. The male opens his own way out of the gall; then he makes an opening into the gall containing the female and fertilizes her. The female later enlarges this same hole and comes out.

The body of the adult female is covered with spines and is black. On examining the wings of the blastophaga, which are also spiny but transparent, it was found that spores, as well as pollen grains, were sticking to them. Insects caught in the vials attached to the eye of caprifigs, as previously described, were found carrying such spores on their wings. Under the moist conditions inside the little vial, the result of transpiration by the fig, these spores were found germinating, and when the insects were left long enough in the vials they became covered with the mycelium of the fungus. A photomicrograph of a portion of the blastophaga wing carrying microconidia of the fungus is shown in plate 15, *a*. It appears reasonable that such

spores are present on other parts of the body of the insect but are not visible against the black background of the insect's body. It is concluded, therefore, that the transmission is "external-mechanical."

In order to determine whether the infection may be also carried by the third method of transmission, viz., internally (mechanically or biologically) the following plan of investigation was outlined.

1. Gall flowers were removed from a caprifig just before the insects issued and were placed on a poured agar plate; some unsterilized, some after sterilization by momentary dipping in 95 per cent alcohol, and others after dipping in mercuric chloride 1:5000 solution, for 1, 2, or 3 minutes, followed by washing in sterile water until the washings were free from chlorine as shown by adding a drop of silver nitrate. To facilitate the handling of such small objects through these steps, the material was kept in sterile Gooch crucibles through all the washings and picked out of the crucible by means of a sterile wire having one end flattened and bent at a right angle.

2. Insects were removed with the aid of sterile needles from their galls, which were sterilized as in (1). The insect and its gall in each case was plated separately.

3. Adults were collected as they were wandering in the cavity after they came out of the gall but before they left the receptacle, and plated, some unsterilized and some sterilized as in (1). After sterilization some insects were crushed and some were not.

Since the infection in the cavity of the fig cannot be determined by other than cultural means, the unsterilized gall flowers from the fig used in each experiment served as an index of infection and a check on the effectiveness of sterilization. Under these circumstances a large number of plates had to be poured from a considerable number of figs in order to secure sufficient data on which to base conclusions. The galls were removed from each receptacle by cutting through the stalk with a sterile knife. They were placed in a sterile Petri dish and mixed, after which they were divided into lots of ten and one lot was used in each experiment. The results showed that sterilization with $HgCl_2$ as described above, was effective. Flowers showing both fungus and bacterial infection when plated unsterilized, were found sterile after sterilization, both when crushed on the plate and when left uncrushed. The purpose of crushing was to facilitate the development of parasites if any were present. The results also showed that the insect is sterile inside the gall, since after external sterilization of infected galls, no more growth was obtained when the gall containing the insect was plated after crushing, or when the insect was removed

from the gall by means of sterilized needles and both insect and gall plated separately. It is possible that the $HgCl_2$ penetrated the tissue of the gall and thus disinfected the insect also, although this is rather doubtful, since a few of the insects were found alive upon extraction from the gall, and these were sterile.

The results from step three of the scheme of attack were not as conclusive as those of steps one and two. Insects picked from inside the cavity of the fig, from the vials (plate 9) and as they were coming out one by one through the eye of the fig were plated, both unsterilized and sterilized, as mentioned previously. When unsterilized insects were plated they usually developed the cryptogamic flora of the fig from which they were issuing. If the fig was sterile or contained the fungus or either of the two bacteria or any combination of the three organisms that constitute the cryptogamic flora of the caprified fig and caprifigs, the same was found to be true of the blastophaga in the majority of cases. Over four hundred insects were used in this phase of the studies. After the insects had become infected no method of sterilization proved completely effective. It has been mentioned before that the body and attachments of the adult insects are covered with spines. Spores lodged among these spines might easily escape killing even when alcohol is used to wet the surface of the body before applying the mercuric chloride solution. It was noticed that the growth on the plates was slow in appearing and not very abundant. In many cases sterilization was effective as many insects were found to be sterile.

From these results it may be safely concluded that the infection is carried externally and mechanically, and that the insect picks up the spores in the cavity of the fig after issuing from the gall and before coming out through the eye of the fig. It seems also probable that there is no internal mechanical or biological transmission. This can be also deduced from our knowledge of the life history of the insect. The egg is laid, according to Grandi(15), inside the ovule of the gall flower between the internal integument and the nucellus. The insect develops inside the walls of the gall, feeding on the endosperm, the female does not come out until the male opens a hole in the gall walls. Unless the infection were deposited along with or carried in the egg, the chances of infection of the insect while still in the gall would be very small.

Caprification Studies.—In caprifying the Calimyrna crop the insectiferous profichi are suspended in strings, baskets, or other containers in the trees and the blastophagas coming out of such profichi

and carrying pollen enter the edible figs and, incidentally, infect them with the endosepsis pathogen if they happen to come out of an infected caprifig. In order actually to prove the connection between infected caprifigs and diseased Calimyrnas the following caprification experiments were carried out at Planada in 1924 and at Davis in 1925. Through the kindness of Mr. W. Arnold of Planada, one hundred and twenty large paper hat bags were placed on Calimyrna twigs, two weeks before any blastophagas issued from caprifigs. In each bag a twig bearing from six to ten figs was enclosed. At caprification time one profichi fig was introduced into each bag from places that were known to be heavily infected. After caprification was over the bags were removed and the figs allowed to ripen. Similarly bags were placed on Adriatic, Kadota, and Black Mission trees in the Forkner Fig Gardens Experimental Plots, and on the Adriatics of the Pasa Rica ranch near Fresno. Caprifigs from the same source as for the Calimyrnas were used. The figs were picked as they ripened, taken into the laboratory and examined for rot. If the figs looked healthy, or were doubtful, plates were poured to determine the presence of infection. No plates were poured from figs showing plainly the symptoms of the rot which developed actively. The results are summarized in table 5.

TABLE 5

PERCENTAGE OF INFECTION OF CAPRIFIED CALIMYRNA, ADRIATIC, KADOTA, AND MISSION FIGS, CAPRIFIED WITH INFECTED CAPRIFIGS

Variety	Total number caprified	Number of infected	Per cent infected
Calimyrna	698	404	57.8
Adriatic	55	42	76.3
Kadota	55	20	36.3
Mission	26	6	18.7

In the experiments of 1925 at Davis, the exact flora of each caprifig was determined by plating. Two hundred and thirty-eight twigs were enclosed in bags. One profichi was introduced in each bag at caprification time. When caprification was over and the bags removed, each caprifig from which the blastophagas had issued was taken to the laboratory and examined. If the symptoms of the disease were evident no plate was poured. If the caprifig looked normal a plate was poured, using the method previously described.

As the edible figs thus caprified were ripening they were taken to the laboratory and carefully examined. The results were according

to expectations. Rather few caprifigs used in caprifying the edible figs in the bags were found to be free from infection, but those that proved to be so produced edible figs that were entirely normal. Specifically, in one instance, the caprifig used in caprifying 16 edible figs on a White San Pedro twig proved to be free from infection. All sixteen figs on this twig were normal. Other twigs on Calimyrna, Adriatic, Black Mission, Bourjassotte Panache, Verdal Longue, and seedling trees, which carried from one to six figs, were also free from infection when caprified with non-infected blastophagas. Not all the figs from twigs caprified with infected insects were diseased, indicating that not all the blastophagas, even from an infected receptacle, carry the infection.

Hand Pollination.—It has been mentioned before that certain varieties of the caprifig (Roeding No. 1, Roeding No. 3, Milco) develope pollen without the stimulus of caprification. Such pollen-iferous but non-insectiferous caprifigs are known as blanks and were found to be free from infection with the endosepsis organism. It was thought that if the pollen from such caprifigs was introduced artificially into Calimyrnas, figs should be developed free from infection. The pollen from blanks was shaken onto a sheet of sterile paper and introduced into sterile glass eye-droppers having one end pulled to a fine capillary. The other end was plugged with cotton and connected with a rubber bulb. The capillary end was introduced into the cavity of the fig and sufficient pollen blown into the cavity to secure pollination. The pollinated figs were protected from caprification before and after pollination by enclosing the twigs in manila paper bags. The pollen and the fig from which it was obtained were plated in order to determine their flora. Green fungi were occasionally found, but no *Fusarium* nor the bacteria described previously.

The results of these experiments were entirely convincing when proper precautions were taken to exclude the insects from both the blank and the pollinated fig. Twenty-five figs were pollinated by this method in 1925 and all proved to be free from disease. The pollen should be collected from the caprifig blanks before the eye of the fig opens because the larva of an unidentified species of thrips may later enter the cavity. Plates were poured repeatedly from blanks and edible figs in order to determine the presence of the thrips larva and its effect on the flora of the fig. Of 95 uncaprified figs examined, the larva was found only in two, and the flora of such figs was never found to parallel the flora of caprified figs. Most of the figs were sterile. Occasionally several types of green fungi that bear no relation to the disease were obtained.

Pathogenesis and Life History of the Pathogen

From the foregoing discussion it is seen that the spores of the fungus are introduced by the blastophaga into the edible fig at a very early stage of growth when the fig is small, green, hard, and ready to be caprified or pollinated. The symptoms of the disease do not appear in the tissues of the fig until it is ripe. In the meantime the fig develops normally, the cavity fills, and the seeds are formed. The question is raised as to what becomes of the spores of the fungus during this interim. Do they germinate and vegetate or remain dormant until the fig tissues can be invaded? Many figs at different stages of development were examined. Occasionally some white, aerial, fungous growth, produced by the endosepsis pathogen was observed in the cavity of unripe figs. This growth invaded very little tissue, but developed mostly on the dead body of the blastophaga which was lodged between the flowers. In many figs the stigmas of certain flowers were much darker than those of others, and this color was very prominent in the flowers surrounding a dead blastophaga. In figs from orchards highly infected, it was found that sometimes the entire cavity where the styles and stigmas come together in the center presented this scorched, dark brick-red pigmentation, although no fungous growth was evident to the naked eye. The styles were removed and examined under the microscope. To facilitate the examination the styles were fixed in absolute alcohol, stained in 2 per cent solution of Magdala red in 85 per cent alcohol, dehydrated in absolute alcohol, cleared in xylol and mounted in balsam. It was found that the withered stigmas were covered with slender hyphae. Plates were poured from figs showing the dark-red stigma, and also from unpigmented ones. The fig was split under aseptic conditions and the entire flower removed by cutting the stalk with a sterile knife. The flowers were placed on poured and cooled Czapek agar plates. It was found that *F. moniliforme* var. *fici* grew regularly from dark-red, orange, or brown-colored stigmas. White or slightly yellow-colored stigmas were sterile. The fungus could be very easily seen on the plates, forming colonies radiating from dark-red stigmas. In one case dark-red and colorless stigmas were selected from the same receptacle and while the former were found infected with the fungus, nothing grew from the latter. Plate 14 (2545 and 2571) illustrates this point. It was found that wherever insects were lodged, as mentioned previously, such brown discolorations could be seen on the

style, the stigma, the stalk, and even on the side of the seed. Fungus hyphae could be seen under the microscope on all of these discolored parts.

Adriatics caprified experimentally were also examined and compared with non-caprified figs. In non-caprified figs of the Adriatic variety the stigma is colorless almost to maturity, when it shrivels and turns yellowish-brown. In caprified figs, immediately after caprification the stigmas turn yellow or yellowish-brown. This discoloration of the stigma is uniform throughout the cavity and cannot be confused with the color of the stigmas showing fungous infection. The latter usually occur in spots, except in extreme cases, and are most abundant near the body of a dead blastophaga. The same is the case in the caprifigs. The fungus was found vegetating on the stigmas until the other tissues could be invaded (plate 15, *b* and *c*).

Rotting tissue of Calimyrnas was fixed in chromacetic solution, washed in water, dehydrated, cleared, infiltrated in paraffin, and sectioned. Various differential stains were tried, as well as Haidenhain's iron-haematoxylin, Delafield's haematoxylin, and Flemming's triple stain. In most sections the disintegration of the tissues was so far advanced that no picture could be obtained of the action of the fungus on the host. However, by making a large number of sections, especially of the meat around the eye, from the margin of watersoaked spots, it was found that the fungus follows at first the vascular system and then invades the cells, growing both intercellularly and intracellularly. Figure 2 and plate 16 illustrate these points.

The life history of the pathogen can now be recapitulated. Infection takes place at caprification time, from spores carried mechanically by the caprifying insect *Blastophaga psenes* L. The spores are usually deposited near or on the flowers about the eye of the fig. The spores germinate readily and the fungus grows slowly on the stigmas of the flowers and the body of the dead insect, until the pulp and the meat commence to ripen and are in turn invaded. The fungus does not attack any part of the tree except the fruit. It does not run into the twigs from the stalk of the fig. Figs are produced at the axils of the leaves of the current year's growth, therefore they could not be infected from old cankers even if such existed. No other source of infection has been found but that of caprification. Within edible figs, therefore, the life cycle of the disease is not completed as the blastophaga is unable to breed in them. The pathogen completes its life cycle in the caprifig paralleling exactly

that of its carrier. The fungus overwinters along with the blastophaga in the mamme figs. In April the adult females carry the spores of the fungus into the profichi where they germinate and grow on the stigma of the gall flowers and the body of the dead insect. In June the adult females come out of the profichi carrying spores of the fungus on their bodies. Some of these females enter Calimyrnas and infect them, others enter mammoni caprifigs, where they carry the infection and lay their eggs. The blastophaga coming out of the mammoni transmit the infection to the mamme in September and the cycle is completed.

Distribution

The disease discussed in this paper has not been reported definitely from other countries. In a series of monographs on the fig industry of the different districts of Italy by Guglielmi(17) on the district of Lecce, Portale(26) on the district of Mistretta, De Rosa(33) on the district of Castro, Siniscalchi(39) on the province of Salermo, and Ferrari(13) on the district of Cosenza, although the diseases of the fig are discussed, no mention is made of internal rot. A gummosis of the fruit of the Dottato (Kadota) is mentioned by Ferrari(13) from the district of Cosenza and discussed in relation to a bacteriosis of the tree. He states that those figs attacked by the disease when nearly ripe show a drop of liquid at the eye first yellow, then reddish, which increases in volume and if the infection is heavy, is exuded. This description corresponds rather to souring, especially since the Dottato is a parthenocarpic variety. No mention of internal rot is made by Trabut(42) or Guillochon(18) from North Africa (Algiers), or Esterlich(12) from Spain in their treatises on the fig. Vallese(44) describes a disease of the fruit in Italy that may possibly be the same as endosepsis. He states that the spoiled fruits ripen without presenting external signs of the disease. The pulp, however, turns pale brown and the sweet juices change to bitter and are extremely disgusting. The most susceptible variety is the red Dottato. Probably this rot is caused by a fermentation of the sugar of the fig started by a bacterium.

Efforts have been made to ascertain whether endosepsis is found in Asia Minor, Algeria, and Greece, where the practice of caprification is in general use. No reference in literature was found. Dried figs from Asia Minor secured by Mr. Condit were examined and plates poured, but without definite results. *Aspergillus* sp. and a few yeasts were the fungi obtained. It was thought that if caprifigs could be

secured from the above mentioned countries in the winter, the flora secured from plating these caprifigs would give an idea regarding the presence of the internal rot in these countries. Permission was obtained from the Federal Horticultural Board and letters were written to scientists and others in the Mediterranean countries. Several responded very kindly and I wish to express my obligation to Dr. Cass A. Reed, of the International College, Smyrna, Dr. L. Trabut, of Algiers; Mr. D. P. Caldis, of Mytilene, Greece; Mr. E. Kefalas, of Samos, Greece; and Mr. C. E. Dickerson, Assistant Trade Commissioner, American Embassy, Athens, Greece, for supplying me with caprifigs.

A number of fungi, mostly of the genus Fusarium, were secured from these caprifigs, but preliminary observations did not disclose these to be the same as *F. moniliforme fici,* the organism responsible for internal rot in California.

SUMMARY

A disease of the Calimyrna and other caprified figs is here described for the first time, and the synonymy and economic importance are discussed. The name endosepsis (internal rot) is proposed.

The disease is caused by *Fusarium moniliforme* var. *fici* n. var., which has been found permeating the diseased tissues, constantly isolated from them, proven to be pathogenic when inoculated and readily reisolated. Strain 93 is considered the type form of the new variety.

The above fungus, together with two bacteria, constitute a typical flora in caprified figs in California.

The organisms constituting this flora are described and their physiological and cultural reactions given. Their taxonomic relations are discussed.

These organisms were found to be transmitted mechanically on the body of the caprifying insect, *Blastophaga psenes* L.

The life history of the pathogen is outlined.

Figs were found to be sterile internally up to the time when they are entered by insects.

ACKNOWLEDGMENTS

The present investigation started in the spring of 1923. The work was carried out for the greater part in Fresno, California, and extensive trips were made into the fig belt of California, comprising that part of the San Joaquin Valley between Modesto and Strathmore. I wish to acknowledge here my indebtedness to Prof. R. E. Smith, under whose direction the whole work was carried on, for valuable suggestions and criticisms, to Prof. W. T. Horne for similar favors, to Mr. W. S. Ballard of the United States Department of Agriculture for the facilities of his laboratory and for valuable assistance and suggestions at all times, and to Mr. I. J. Condit for many favors and for several photographs appearing in this paper.

LITERATURE CITED

(1) Society of American Bacteriologists
1923. Bergey's manual of determinative bacteriology, 1–442. Williams and Wilkins Co., Baltimore.

(2) Caldis, P. D.
1925. A rot of the Smyrna fig in California. Sience, n.s., **62**:161–162.

(3) Chen, C. C.
1920. Internal fungous parasites of agricultural seeds. Maryland Agr. Exp. Sta. Bul. **240**:81–110.

(4) Condit, I. J.
1921. Why our trees are dying. Farm Bureau Review, **1**:15.

(5) Condit, I. J.
1922. Caprifigs and caprification. California Agr. Exp. Sta. Bul. **319**: 341–377.

(6) Condit, I. J.
1919. Die-back of the fig in California. Fig and Olive Jour., **4**:11–12.

(7) Coons, G. H.
1916. Factors involved in the growth and the pycnidium formation of *Plenodomus fuscomaculans*. Jour. Agr. Res., **5**:713–769.

(8) Cotte, J., et A. Reynier
1923. La dioecie du figuier et *Blastophaga psenes* L. Compt. Rend. Soc. Biol., **88**:500–502.

(9) Edgerton, C. W.
1911. Diseases of the fig tree and fruit. Louisiana Agr. Exp. Sta. Bul. **126**:1–20.

(10) Eisen, Gustav
1896. Biological studies on figs, caprifigs and caprification. Proc. California Acad. Sci., II, **5**:897–1003.

(11) Eisen, Gustav
1901. The fig: its history, culture, and curing. U. S. Dept. Agr., Div. Pom. Bul. **9**:1–317.

(12) Esterlich, Pedro
1915. La higuera y su coltivo en España, 1–228. Barcelona.

(13) Ferrari, E.
1912. La coltivazione del fico nel Circondario di Paola (Consenza) memoria monografica. Ann. R. Staz. Agrum. Frutticol., **1**: 141–177.

(14) Gould, H. P.
1923. Fig growing in the South Atlantic and Gulf States. U. S. Dept. Agr. Farmers' Bul. **1031**:1–47.

(15) Grandi, G.
1920. Studio morfologico e biologico della *Blastophaga psenes* L., 1–144. Portici.

(16) Grandi, G.
1923. Gli insetti dei caprifichi. Riv. Biol., **5**:1–26.

(17) Guglielmi, G.
1908. Coltivazione industriale del fico nel leccese. Memoria monografica. Bol. Arboric. Ital., **4**:11–17, 57–64, 114–125, 152–156.

(18) Guillochon, G.
1913. Traito pratique d'horticulture pour le Nord d'Afrique, 263–270. Tunis.

(19) Hodgson, R. W.
1918. A black rot of figs. Phytopath., **8**:545–546.

(20) McWhorter, F. P.
1926. A simple method for making counts of small spores. Science, n.s., **63**:211–212.

(21) Matz, J.
1918. Some diseases of the fig. Florida Agr. Exp. Sta. Bul., **149**:1–10.

(22) Morris, H. E., and Grace Nutting
1923. Identification of certain species of *Fusaria* isolated from potato tubers in Montana. Jour. Agr. Res., **24**:339–364.

(23) Phillips, E. H.
1922. Some fig diseases. Phytopath., **12**:107.

(24) Phillips, E. H., E. H. Smith, and R. E. Smith
1925. Fig smut. California Agr. Exp. Sta. Bul., **387**:1–38.

(25) Pierce, N. B.
1893. Investigations of the special agent in California. U. S. Dept. Agr., Secy. Rept., **1892**:238–241.

(26) Portale, F.
1910. La coltivazione del fico nel circondario di Mistretta. Memoria monografica. Boll. Arbor. Ital., **6**:1–53.

(27) Porter, C. L.
1924. Concerning the characters of certain fungi as exhibited by their growth in the presence of other fungi. Am. Jour. Bot., **11**: 168–188.

(28) Potts, A. T.
1917. The fig in Texas. Texas Agr. Exp. Sta. Bul., **208**:1–41.

(29) Rand, F. V., and W. D. Pierce
1920. A coördination of our knowledge of insect transmission in plant and animal diseases. Phytopath., **10**:189–231.

(30) Rettger, L. F., and J. L. Sherrick
1911. Studies on bacterial variation. Jour. Med. Res., **24**:265–284.

(31) Rixford, G. P.
1918. Smyrna fig culture. U. S. Dept. Agr. Bul. 732:1–43.

(32) Roeding, G. E.
1903. The Smyrna fig at home and abroad, 1–87. Fresno, Calif.

(33) Rosa, Francesco, de
1911. Di alcuni fichi Salentini. Atti R. Inst. Incorr. Napoli, VI, 9: 1–36.

(34) Scheurlen
1896. Geschichtliche und experimentelle studien über den prodigiosus. Archiv. für Hygiene, 26:1–13.

(35) Sheldon, J. L.
1903. A corn mold, *Fusarium moniliforme* n. sp. Nebraska Agr. Exp. Sta. Rept., 17:23–32.

(36) Sherbakoff, C. D.
1915. Fusaria of potatoes. Cornell Univ. Agr. Exp. Sta. Mem. 6:97–269.

(37) Sideris, C. P.
1925. Studies on the behavior of *Fusarium cromiophthoron* Sid. in carbohydrates, glucosides, proteins and various decoctions, with discussion on the "isometabolic point" of substances. Phytopath., 15:129–145.

(38) Sideris, C. P.
1925. The rôle of the hydrogen-ion concentration on the development of pigment in Fusaria. Jour. Agr. Res., 30:1011–1019.

(39) Siniscalchi, A.
1911. La coltivazione del fico nel Gilento (Prov. de Salermo). Memoria monografica. Boll. Arbor. Ital., 7:25–54.

(40) Smith, E. H., and E. H. Phillips
1922. Studies of the so-called "smut" of white fig varieties. Monthly Bul. California State Dept. Agr., 11:755–758.

(41) Stevens, F. L., and G. Hall
1909. A new anthracnose of figs. North Carolina Agr. Exp. Sta. Rept., 32:86–89.

(42) Trabut, L.
1923. Ennemis et maladies du figuier. Bul. Agric. Afr. Nord., 29: 117–124:

(43) Vallese, Ferdinando
1904. La caprificazione in Terra d'Otranto, 1–68. Lecce.

(44) Vallese, Ferdinando
1909. In fico. Biblioteca d'agricoltura e industrie affini, 1–374. Catania.

(45) Wineland, G. O.
1924. An ascigerous stage and synonymy for *Fusarium moniliforme*. Jour. Agr. Res., 28:909–933.

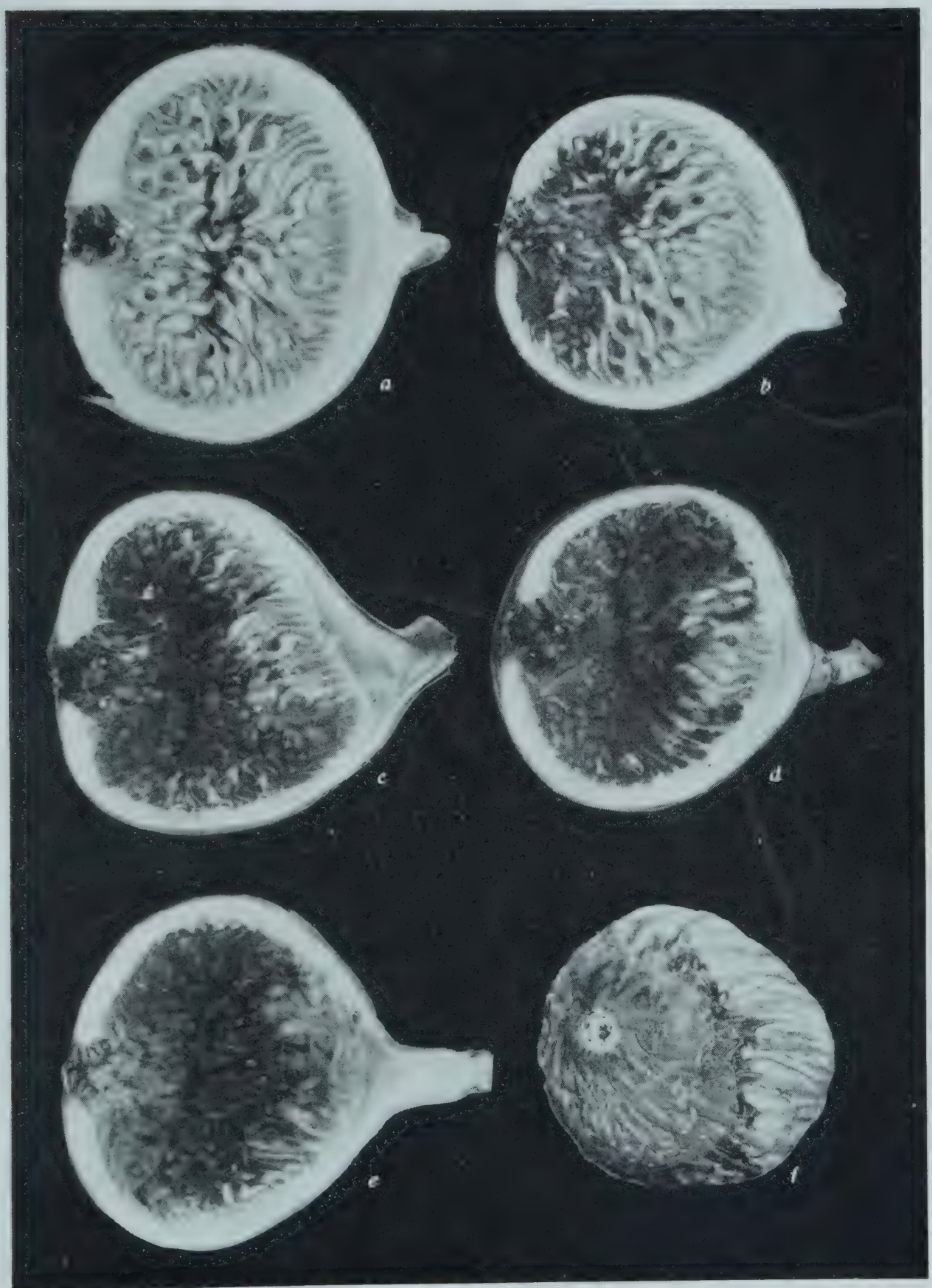
(46) Wolff
1909. Modifications and mutants of *B. prodigiosus* and other Schizomycetes produced experimentally. Zeit. Indukt. Abst. Vererb., 2:90.

(47) Wollenweber, H. E., C. D. Sherbakoff, O. A. Reinking, H. Johann, and A. A. Bailey
1925. Fundamentals for taxonomic studies of Fusarium. Jour. Agr. Res., 30:833–843.

(48) Wollenweber, H. E., and O. A. Reinking
1925. Aliquot Fusaria tropicalia nova vel revisa. Phytopath., 15:155–170.

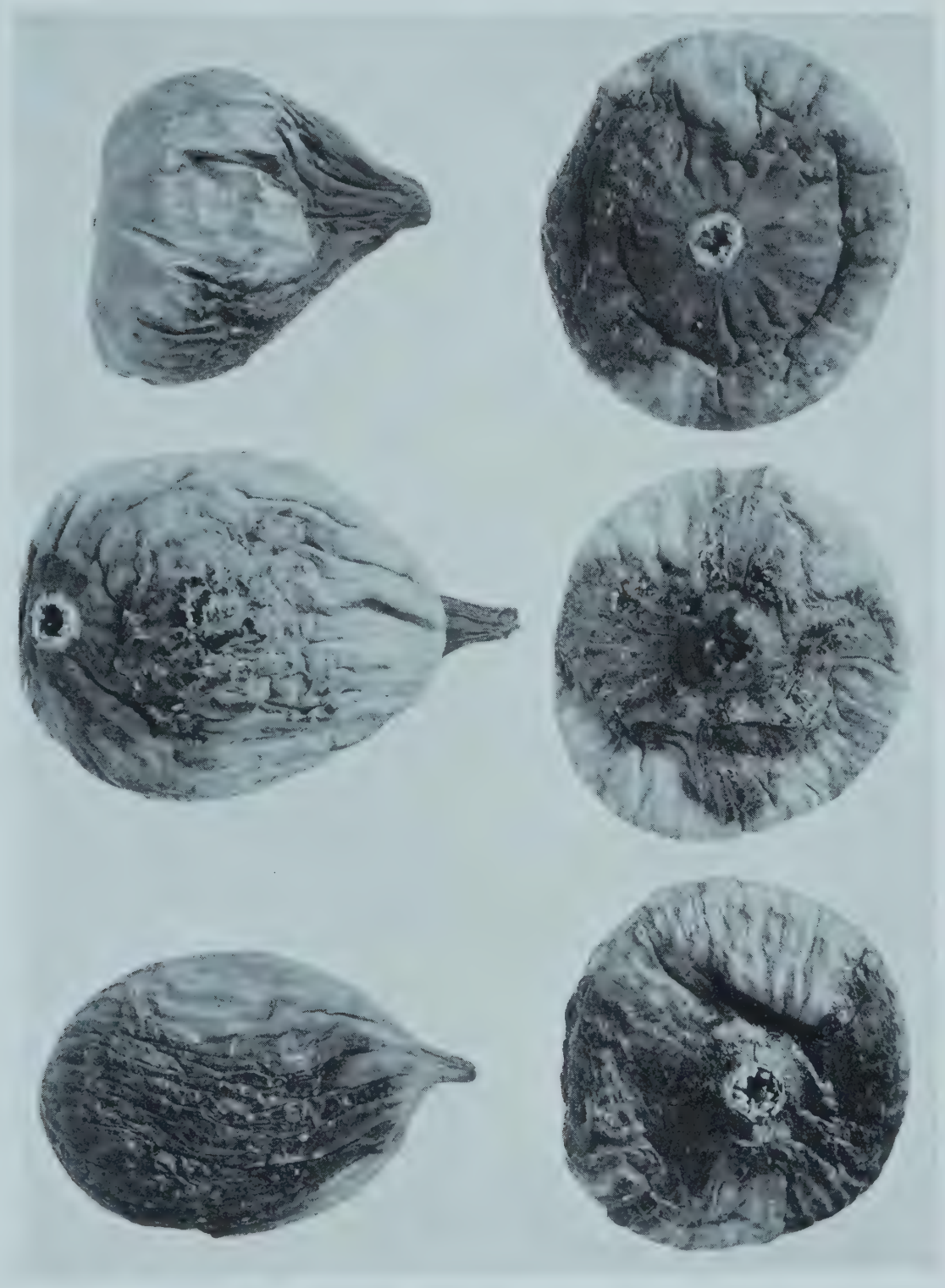
PLATES 1-16

Transmitted September, 1926.



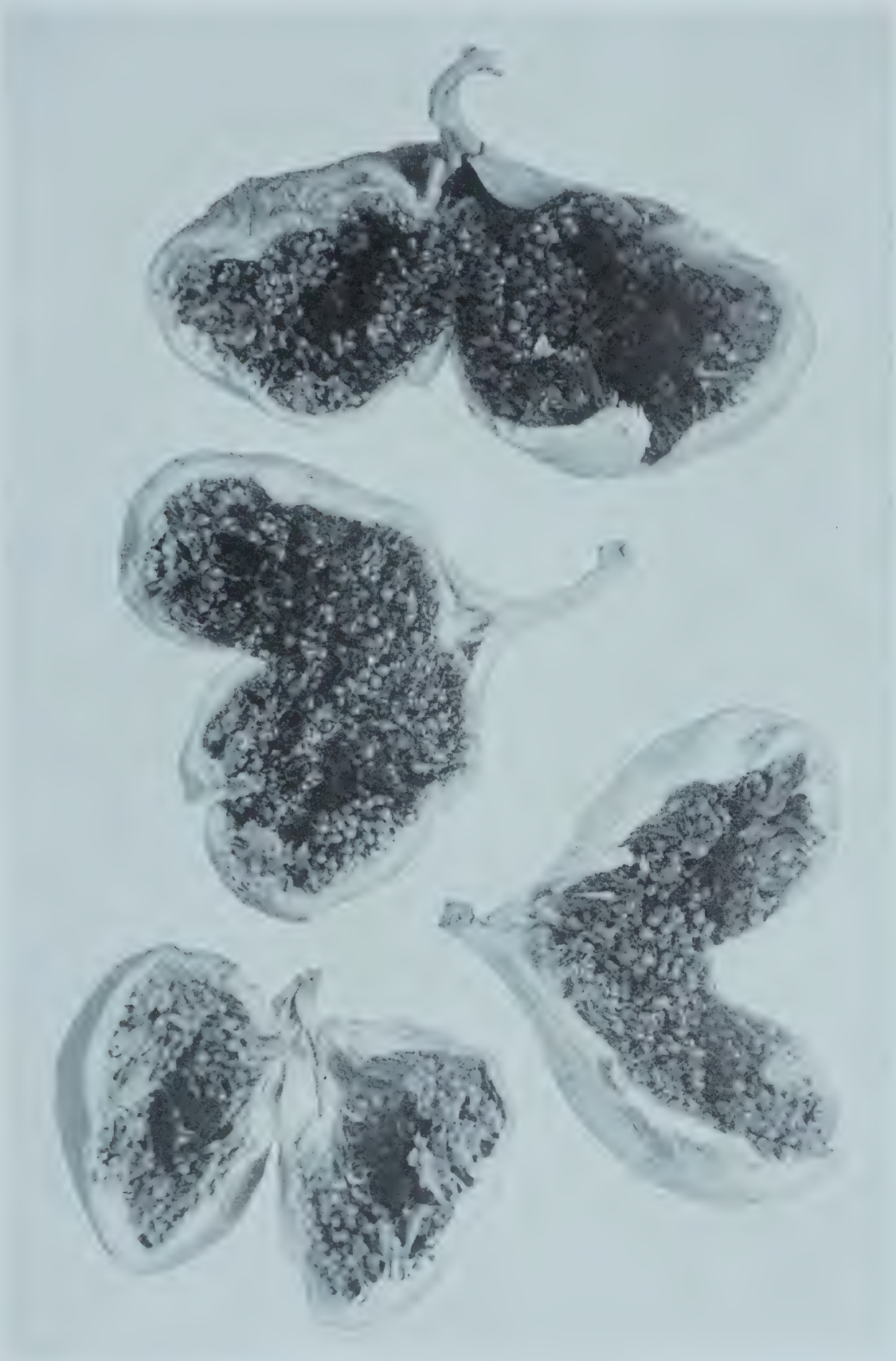
Calimyrna figs at stage of maturity ready for picking for fresh shipment or canning, showing progressive internal symptoms of endosepsis. External appearance normal in the first five. *a*, Normal fig. *b*, Flowers around the eye diseased. *c*, Three-fourths of the pulp diseased. *d*, Seven-eighths of the pulp diseased. *e*, Entire pulp diseased. *f*, External symptoms. Meat invaded.

Calimyrna figs showing external symptoms of endosepsis.

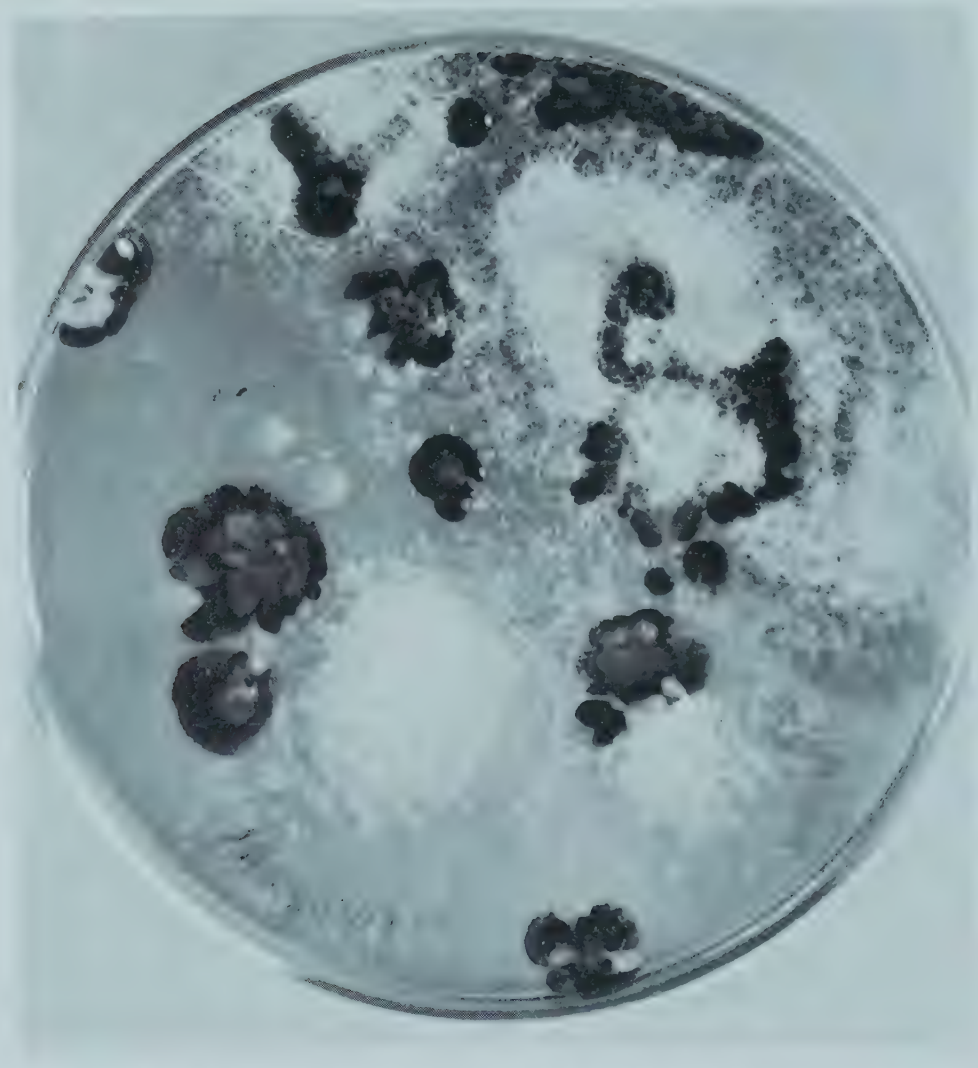


Dried Calimyrna figs showing external symptoms of endosepsis. The dark watersoaked spots are pink, red, or purple in color. Notice the small drop of gum at the eye of one fig.

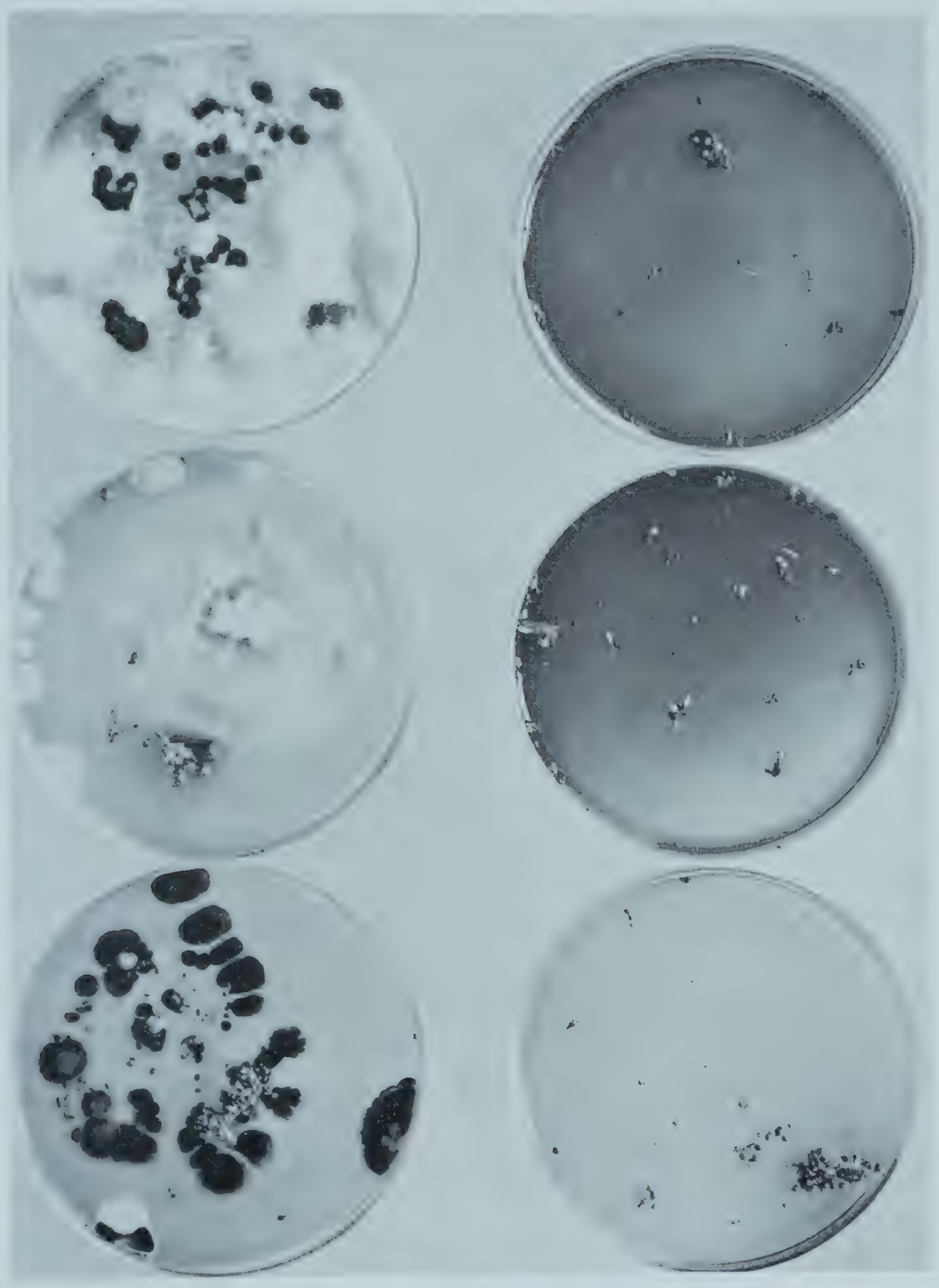
Dried Calimyrna figs of practically normal appearance but internally slightly affected with endosepsis.



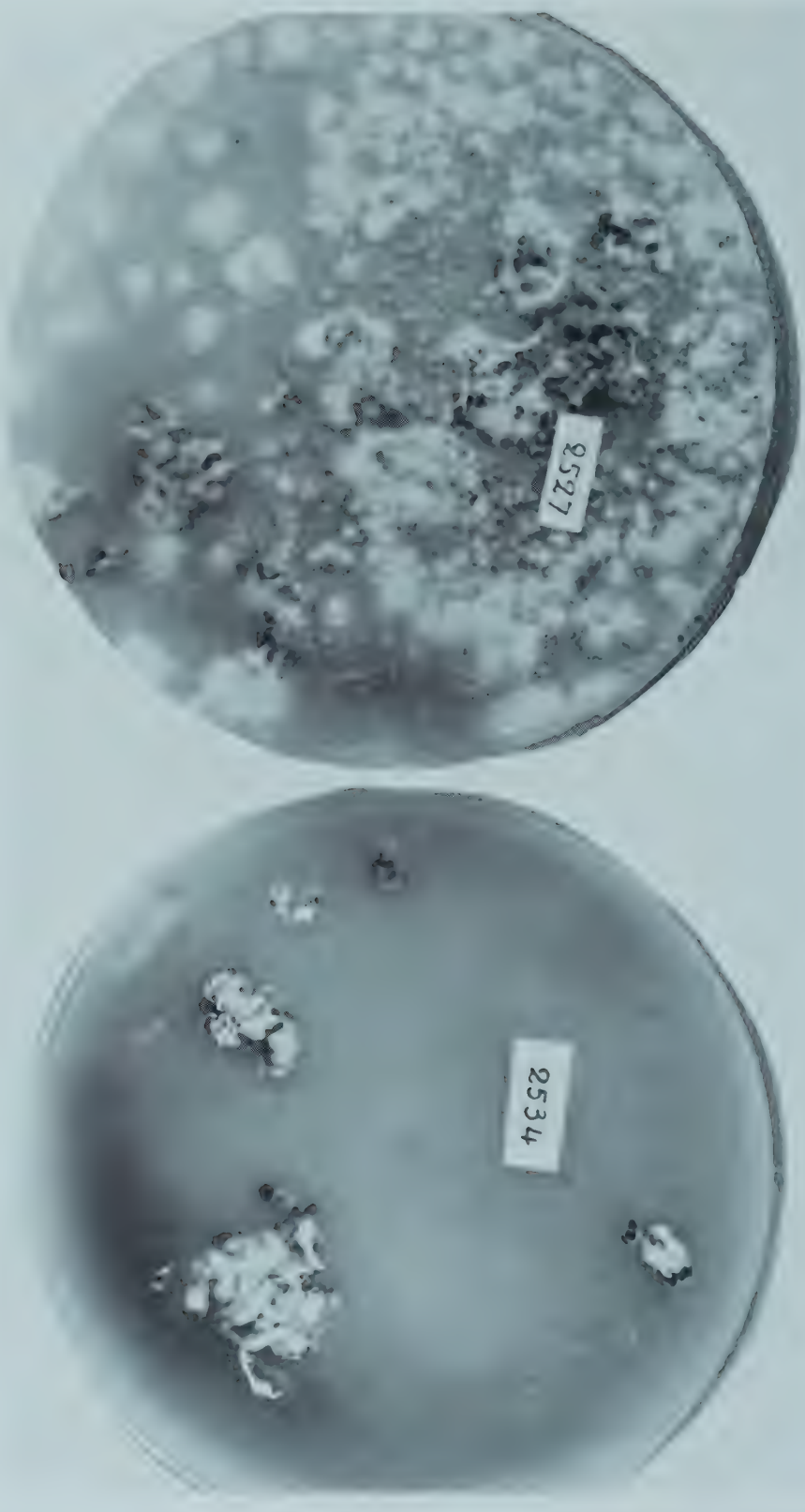
Interior of dried figs shown in plate 4, slightly affected with endosepsis. The pulp is dry and "seedy" and has a peculiar flavor.



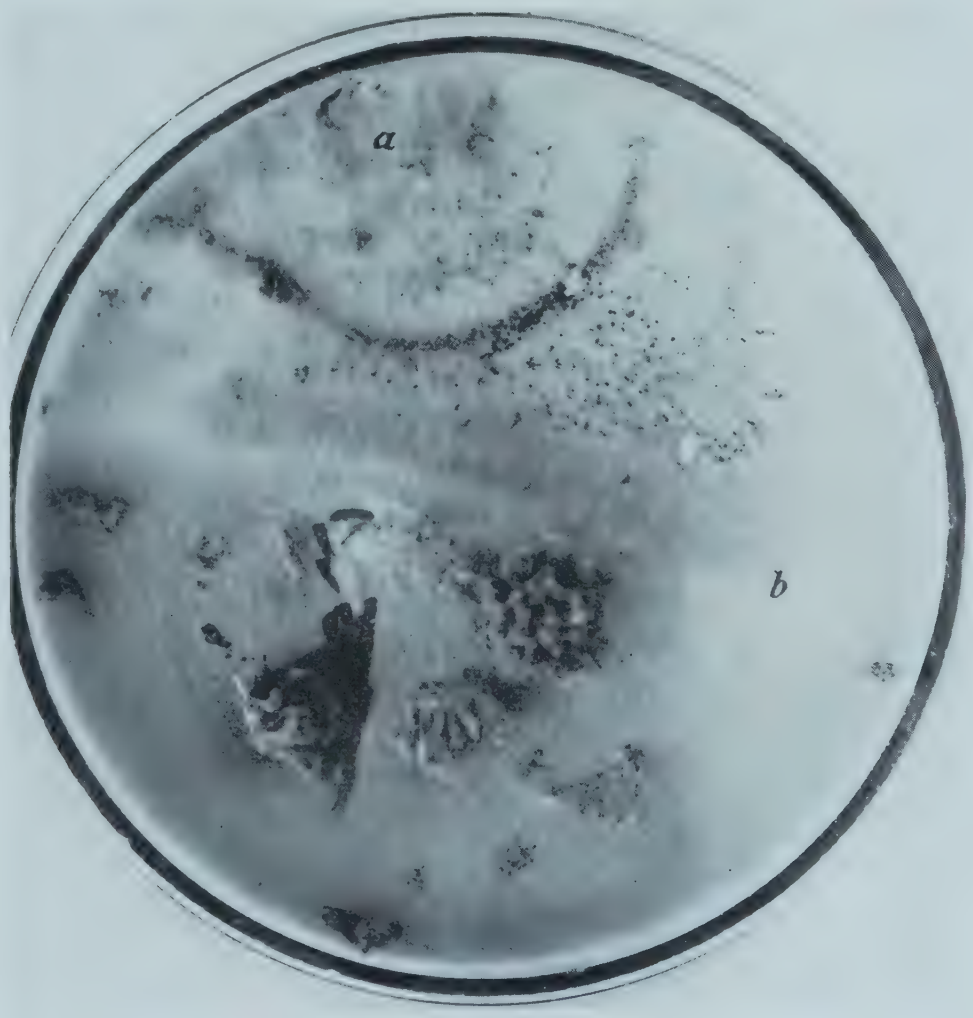
Typical flora of a caprified fig, showing the three characteristic organisms.



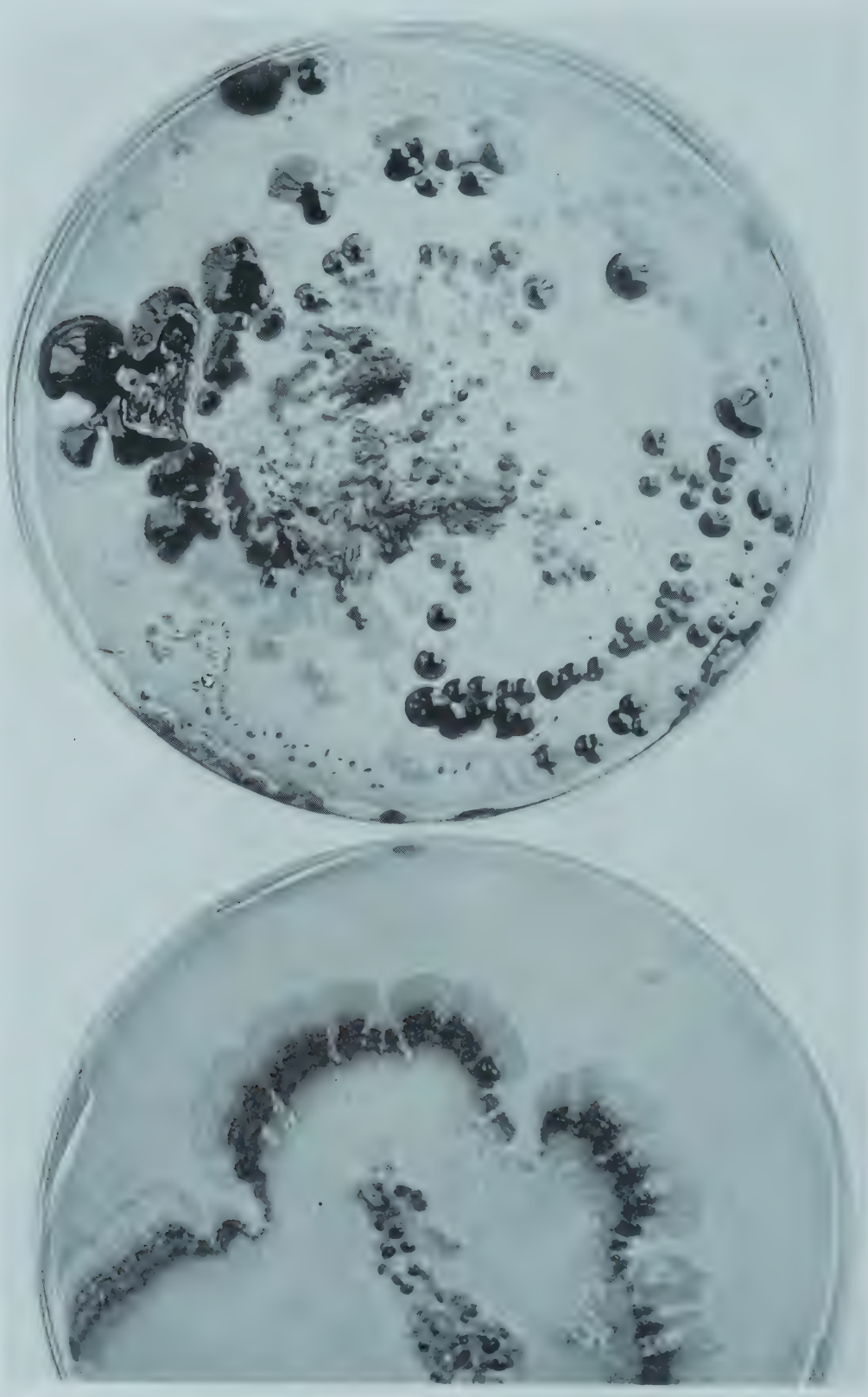
Left, typical plates from unripe, caprified, Calimyrna figs. Right, plates poured from uncaprified, unripe, Mission, Kadota, and Adriatic figs; sterile.



2527, flora of a caprified, unripe Adriatic. 2534, flora of a non-caprified Adriatic; sterile.

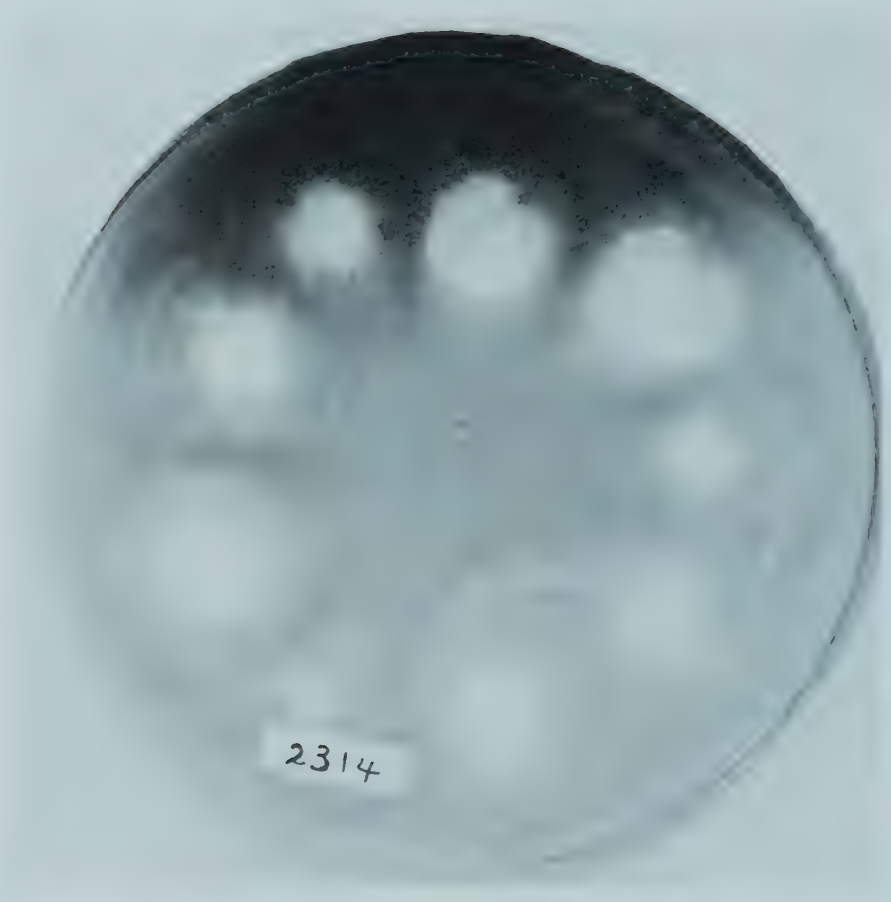


Above, method of catching blastophaga as they issue from caprifigs. Below, formation of perithecia in *F. moniliforme fici.*



Two plates illustrating the growth of the red chromogenic bacterium obtained regularly from caprified figs.

Caprifigs at the time of infection. *a*, Profichi. *b*, Mamme.

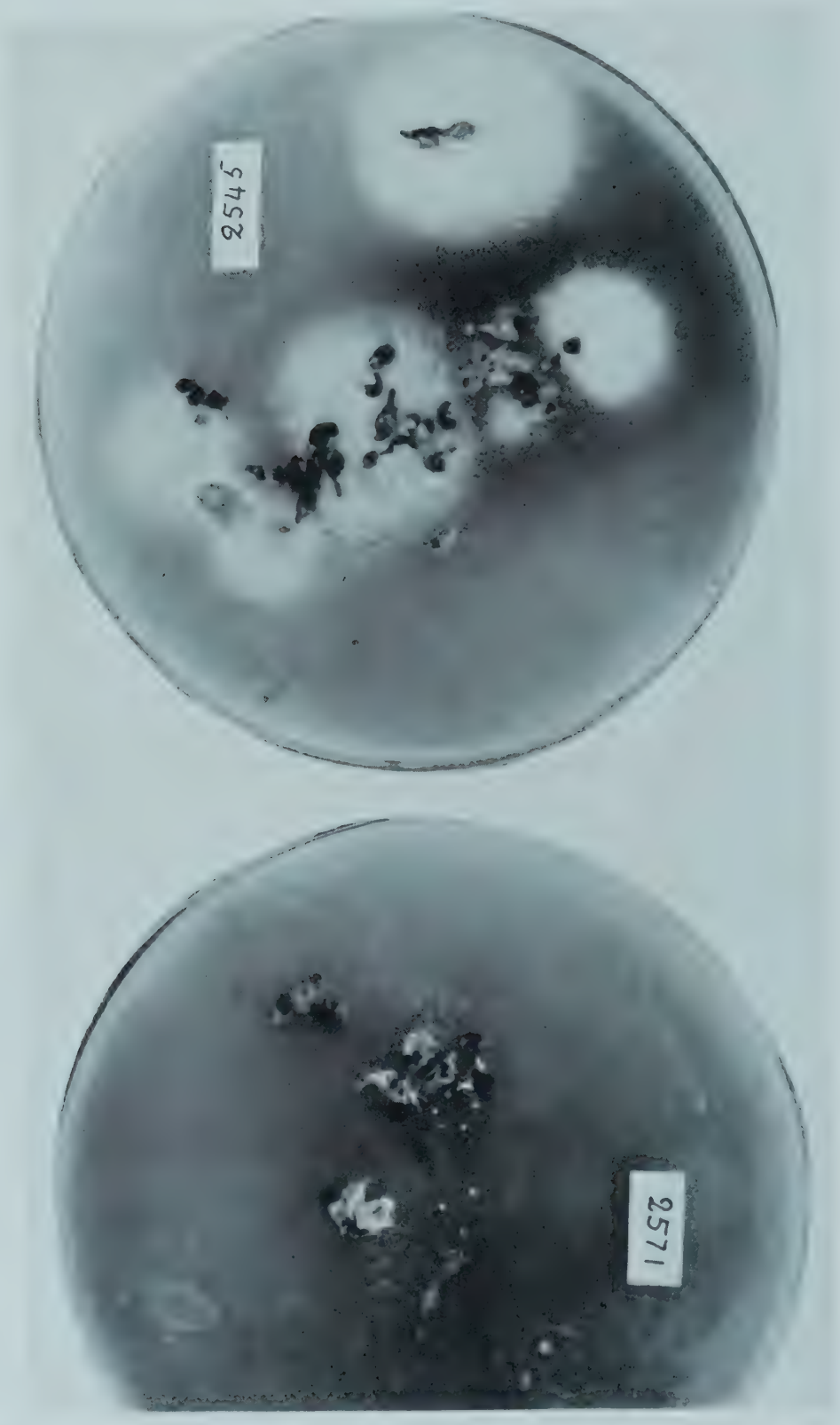


2387, galls picked from the cavity of an infected protichi. Notice the typical growth of *F. moniliforme fici.*

2314, blastophaga caught in the traps shown in plate 9 were placed on this poured agar plate. Notice the growth from each. The central puncture represents the control.

2310. *a*, Growth from blastophaga taken from a fig infected with red bacterium and white fungus. *b*, Growth from blastophaga taken from a fig infected only with white fungus.

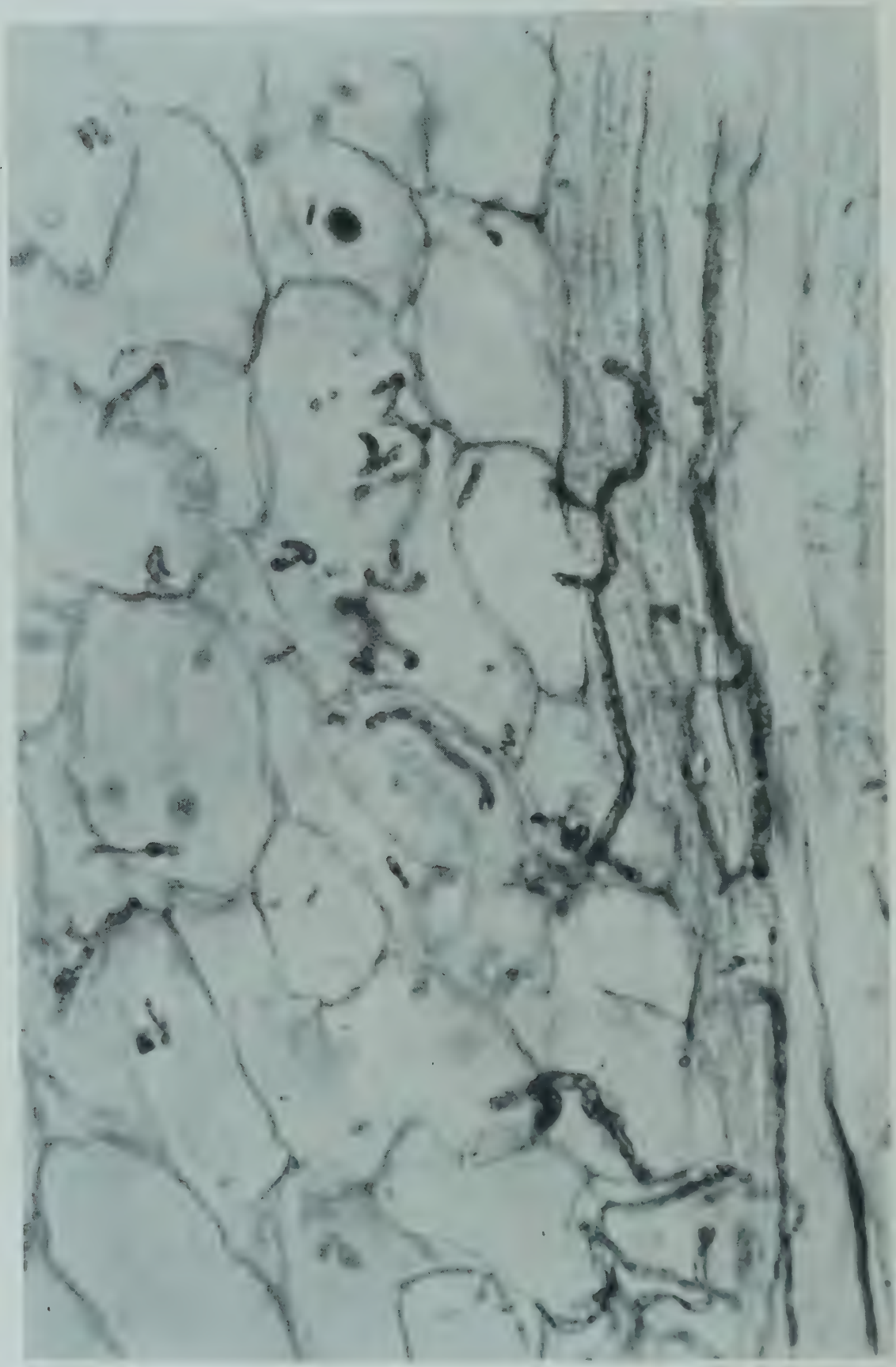
2389. *a*, Growth from galls from a fig infected with white bacteria. *b*, Growth from galls taken from a fig infected with red bacteria.



2545, plate poured from flowers showing a brick red colored stigma. Notice the fungous colonies.

2571, plate poured from flowers showing a light yellow colored stigma. Notice the absence of fungus.

a, Part of the wing of a blastophaga, showing the spores of the fungus. *b*, Profichi fig, showing growth of the fungus in the cavity. *c*, Stigma of infected fig flower, showing fungus.



Photomicrograph of a section of ripe Calimyrna meat tissue, showing intercellular and intracellular penetration of the fungous hyphae.

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SOME FACTORS INFLUENCING THE ROOTING OF VINE CUTTINGS

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A perfect "stand" is rarely obtained in rooting vine cuttings. The difficulty of securing a high percentage of rootings[2] varies greatly with the varieties of the different species and their hybrids. Some of the best phylloxera-resistant stocks are rooted with great difficulty. When the importance of propagating by cuttings and the difficulties of rooting are considered, the value of any treatments or methods of handling which will increase the proportion of cuttings that root or which will improve the quality of the rootings produced is obvious.

The objects of this investigation were to determine the influence on the number of cuttings that rooted and on the quality of the rootings produced of (1) the starch content of the cutting, as indicated by the iodine test, (2) the time of planting, and (3) treatment with oxidizing agents.

THE STARCH CONTENT OF CUTTINGS AS INDICATED BY THE IODINE TEST

The iodine test as an indication of the stored reserve foods of cuttings has been successfuly applied by some of the leading nurserymen of Europe in selecting stocks for grafting. This test, however, is not used in the selection of cuttings for planting either in the vine

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[2] Rootings, as used in this paper, refers to cuttings which have developed roots and grown for one year in the nursery.

nursery or directly in the field in California. Since the varied climatic conditions of California greatly affect the nutrition of the canes through their influence on the nature of the growth of the vines, the iodine test may be of value in eliminating cuttings which do not possess adequate reserve foods.

In testing the relation of starch content to rooting, as indicated by the iodine test, cuttings of Sultanina were collected from several of the grape-growing sections of the state. The cuttings were made in January and shipped by express to Davis, where they were held in sand in a cool sand pit until the middle of February, when the tests were carried out.

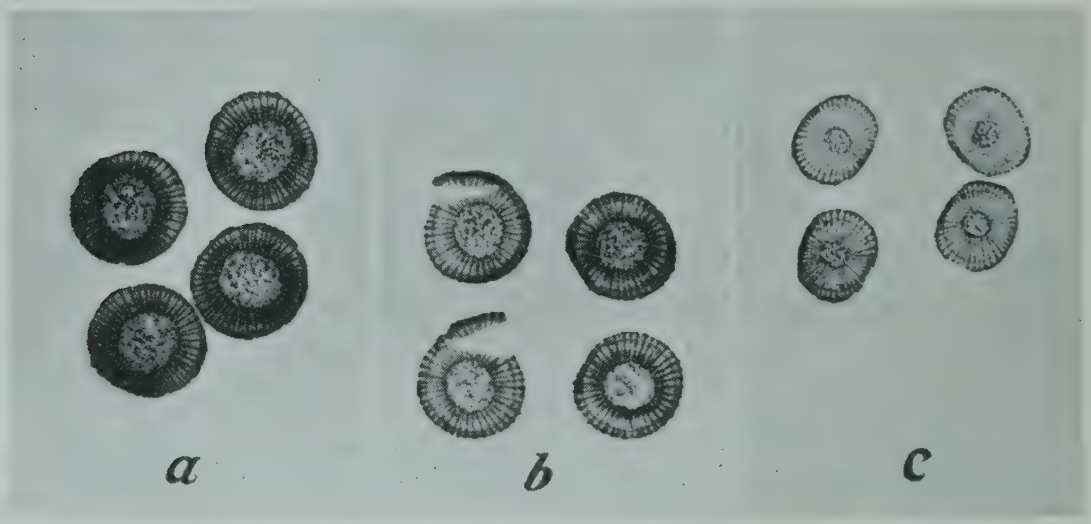

Fig. 1. The different degrees of intensity of staining with iodine corresponding to the starch content of the cuttings in the several groups. *a*, Group 1. *b*, Group 2. *c*, Group 3.

In making the tests, freshly cut ends of two hundred cuttings of each lot were immersed for one minute in a 0.2 per cent solution of iodine in potassium iodide. By means of this uniform exposure to the iodine solution, it was possible as a result of the different degrees of intensity of staining obtained to divide the cuttings into the following groups:

Group 1: Cuttings which showed deep staining throughout the wood and very dark staining in the medullary rays (fig. 1, A).

Group 2: Cuttings which showed only faint staining in the wood near the cortex, slight staining of the wood adjacent to the pith, and deep staining in the medullary rays (fig. 1, B).

Group 3: Cuttings which showed no staining in the wood near the cortex, faint staining of the wood adjacent to the pith, and well defined staining in the medullary rays (fig. 1, C).

Although it was impossible to make a very rigid classification of cuttings by this method of selection, the grouping as indicated above appears to be serviceable for practical purposes. The results, at least, seem to indicate that the slight overlapping of the groups which of necessity occurred, was not sufficient to greatly influence the results. The percentage of rootings obtained under the different groups is given in table 1.

TABLE 1

THE INFLUENCE OF THE STARCH CONTENT ON THE PERCENTAGE AND THE VIGOR OF THE ROOTINGS PRODUCED

	Cuttings with high starch content (Group 1)	Cuttings with medium starch content (Group 2)	Cuttings with low starch content (Group 3)
Per cent rooted	62.5	35.3	16.9
Per cent of vigorous rootings produced	30.0	9.3	1.8

The figures of table 1, which are percentages of the number of cuttings planted, indicate that there is a more or less direct relationship between the starch content of the cuttings and the number of rootings produced. The data further indicate that the vigor of the rootings produced is also closely correlated with the starch content.

TABLE 2

THE RELATION OF REDUCING SUBSTANCES AND STARCH CONTENT TO THE NUMBER AND VIGOR OF THE ROOTINGS PRODUCED UNDER THE SEVERAL GROUPS

	Cuttings with high starch content (Group 1)	Cuttings with medium starch content (Group 2)	Cuttings with low starch content (Group 3)
Per cent of reducing substances	2.27	2.30	2.12
Per cent of starch	5.68	3.72	2.56
Per cent of cuttings rooted	85	42.5	23.5
Per cent of vigorous rootings	53.1	17.0	5.1

As a further check on the reliability of the iodine test, analyses were made of a considerable number of cuttings of the different groups. These analyses, together with the percentage and vigor of the rootings produced, are given in table 2.

The figures of table 2 indicate that the iodine test is relatively accurate and that it may, at least for practical purposes, serve as a means of determining the storage of reserve foods of cuttings. The

figures also emphasize the relation of the starch content to the viability of the cuttings. There seems to be no relation between the starch content and the amount of reducing substances, since the latter substances are present in about the same amount in each of the groups of cuttings.

It is also of interest to note that the vigor of the rootings produced was closely correlated with their composition. Although the rootings of the cuttings lower in starch had a greater space in the nursery row, owing to a greater mortality, this advantage did not enable them to equal the vigor of the rootings of the cuttings of higher starch content. This difference in mortality and its influence on spacing for groups 2 and 3 is shown in figure 2.

Fig. 2. The relation of starch content to the mortality of cuttings in the nursery and the effect of the death of cuttings on the spacing of the remaining rootings. *a*, Group 2. *b*, Group 3.

TIME OF PLANTING

During the seasons of 1924 and 1925, lots of two hundred cuttings each were planted at varying intervals, from December of the previous year to May. These plantings were made to determine, if possible, the influence of the time of planting on the number and vigor of rootings produced. The results obtained during the two seasons are listed in table 3.

The figures of table 3 show a considerable advantage in favor of the earlier plantings with regard to both the total number of rootings produced and the quality of the rootings. All cuttings planted after the middle of March were very unsatisfactory. The falling off in

TABLE 3

THE EFFECT OF TIME OF PLANTING ON THE NUMBER AND VIGOR OF ROOTINGS
(The percentage of rootings produced by the earliest planting is taken as 100)

Variety	Date of planting	Per cent of cuttings that rooted	Per cent of vigorous rootings produced
Alicante Bouschet	Dec. 20, 1923	100	67
	Jan. 16, 1924	90	58
	Feb. 18, 1924	89	49
	Mar. 20, 1924	53	40
	Apr. 15, 1924	38	31
	May 15, 1924	46	12
Sultanina	Dec. 31, 1924	100	77
	Feb. 2, 1925	82	59
	May 3, 1925	24	6
Muscat	Dec. 31, 1924	100	65
	Feb. 2, 1925	67	30
	May 3, 1925	35	8

both the quality and quantity of rootings produced, however, is more or less uniform from the earliest to the latest plantings.

The figures of table 4 and the illustrations of figure 3 indicate why earlier planting gave the better quality and probably also the greater percentage of rootings. These data show that the cuttings of the earlier planting started root development much in advance of those of the late plantings. This not only tended to extend the growing season but also gave the roots a start before the buds pushed.

TABLE 4

THE CONDITION OF ROOT AND TOP DEVELOPMENT OF ALICANTE BOUSCHET CUTTINGS OF THE VARIOUS PLANTINGS ON APRIL 15, 1924

Date of planting	Per cent of cuttings showing root development	Average number of roots per cutting	Average length of the individual roots *cm.*	Per cent of cuttings showing top development
Dec. 20, 1923	60	4.5	8.9	5
Jan. 16, 1924	50	3.4	8.3	7
Feb. 18, 1924	40	3.0	.4	3
Mar. 20, 1924	20	2.5	.1	0
Apr. 15, 1924	0	0	0	0
May 15, 1924	0	0	0	0

TREATMENT WITH CHEMICAL COMPOUNDS THAT EXERCISE STIMULATORY EFFECTS

In the tests of chemical treatments, an attempt was made to improve the rooting of grape cuttings by utilizing the findings of Curtis(2) and others, who have shown that various chemicals, especi-

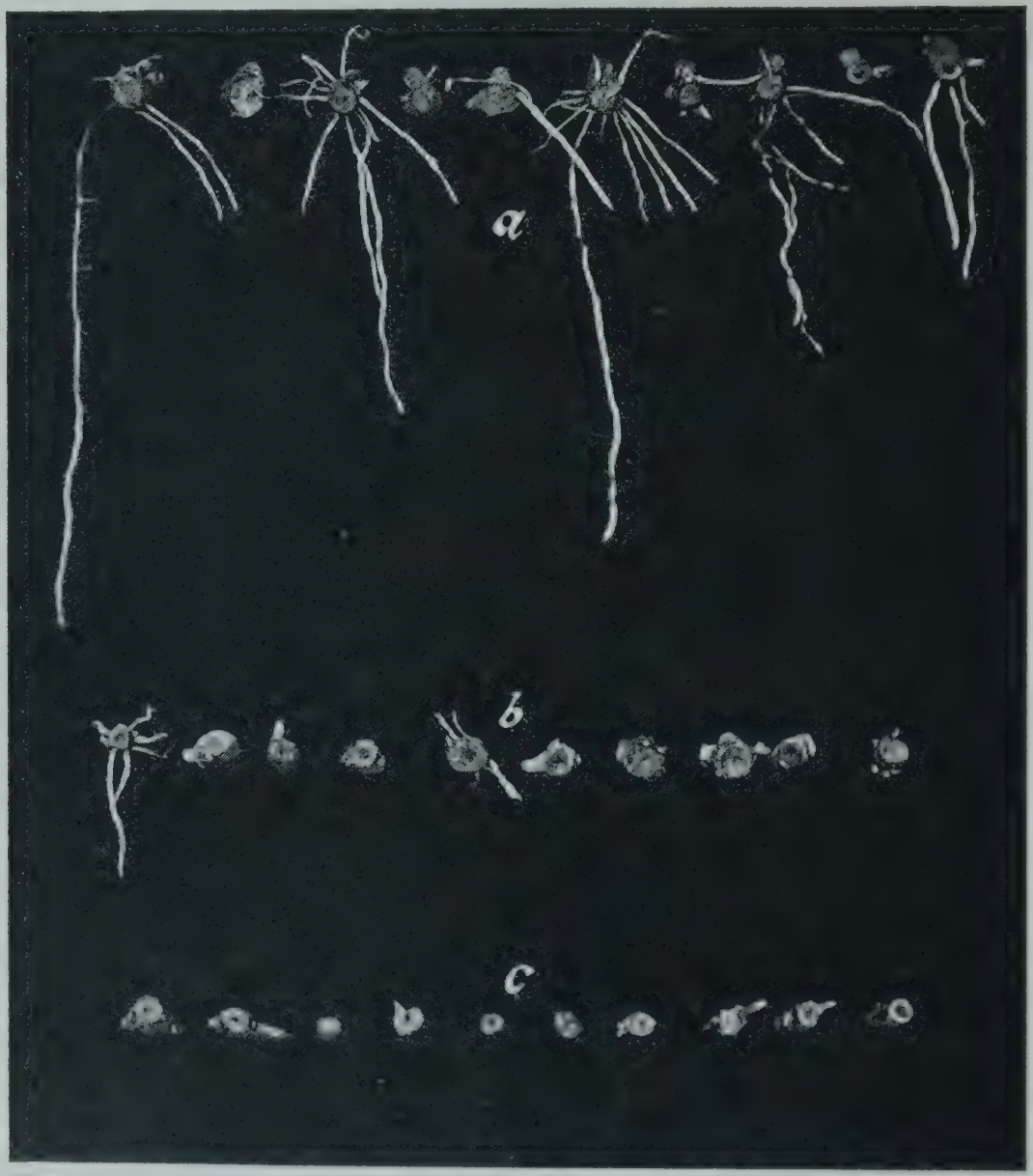

Fig. 3. The influence of the time of planting on the stage of root development on April 15. *a*, Planted December 20. *b*, Planted February 18. *c*, Planted March 20.

ally oxidizing reagents, stimulate root growth. In these tests, cuttings were subjected to continuous contact or to preliminary treatments of short duration in solutions of the various compounds in distilled water. In the continuous treatments, the cuttings were rooted in flasks containing the solution. In the limited treatments, the cuttings

were left in the solution in most cases for forty hours. In all of the treatments about one inch of the base of the cuttings was immersed in the solution.

In a few of the earlier of these tests, twenty cuttings were used in each lot; however, in the majority of the trials, fifty cuttings were used. Glass jars placed on the laboratory bench served as containers for the cuttings. For callusing in sand, the cuttings were reversed in a sand pit with a southern exposure which was covered with glass. The temperature was not controlled. However, where comparisons are made between lots treated with different compounds, the cuttings were all treated as nearly alike as possible with respect to exposure to light and temperature.

In the comparisons, the average percentage of rootings, the vigor of the rootings, the average total length of top and root growth (for the newly rooted cutting), and the diameter of the roots for the one-year-old rootings are used. The length of individual roots and shoots varied too greatly for comparisons of these to be consistent.

Data have been collected on the effect of the duration of treatment and of the concentration of solution of the various compounds together with their influence on the rate of root and callus formation and on the number and vigor of the rootings produced.

Concentration of Solution.—In tests of the effect of concentration of solution twenty cuttings were treated for 24 hours in each of the several concentrations of solution of the several different compounds. After treatment, the cuttings were callused in sand for 14 days. The results of these tests are given in table 5.

The figures of table 5 indicate that cuttings, in the stage of dormancy, of the Alicante Bouschet (a variety of *Vitis vinifera*) cuttings used, are tolerant of a considerable range in the concentration of the reagents without loss of the stimulating effect. This tolerance is a factor of much importance, if this form of root stimulation is to be applied in practice where an accurate control of the concentration of the reagents is difficult.

The concentrations giving the greatest stimulations were .001 to .0001 mol. solutions of $MnSO_4$, $Mn_2(SO_4)_3$, $K_3Fe(CN)_6$, and iodine; .01 to .001 mol. solutions of MnO_2, $FeCl_3$, and Na_2O_2, and .1 to .05 mol. solutions of H_2O_2 and $KMnO_4$. The greatest concentrations of solution in these tests, except for the H_2O_2 and $FeCl_3$, were approaching the maximum concentration that could be used without injury, since the root growth under these was little or no better than that of the check.

TABLE 5

THE INFLUENCE OF CONCENTRATION OF SOLUTION OF THE SEVERAL COMPOUNDS ON THE FORMATION OF ROOTS ON ALICANTE BOUSCHET CUTTINGS

Reagent	Mol. concentration	Per cent of cuttings rooted	Average total root length per cutting
Water (check)	Distilled	30	4.2
$KMnO_4$	.05	70	9.7
	.1	100	32.1
	.2	40	6.5
MnO_2	.001	90	52.2
	.01	95	59.2
	.1	70	35.7
	.2	40	16.0
Na_2O_2	.001	95	72.6
	.01	70	47.7
	.05	35	5.9
	.1	30	2.4
$FeCl_3$	.0001	40	11.6
	.001	65	68.8
	.01	75	47.6
$MnSO_4$	.0001	85	81.3
	.001	100	112.3
	.01	75	47.6
$Mn_2(SO_4)_3$	.0001	65	21.1
	.001	65	12.5
	.01	35	8.7
H_2O_2	.001	55	32.6
	.01	40	19.5
	.1	75	45.0
	.5	90	58.0
$K_3Fe(CN)_6$	.0001	90	16.0
	.001	95	28.8
	.01	60	6.8
$K_4Fe(CN)_6+3H_2O$	.0001	35	6.7
	.001	25	4.2
I	.0001	80	87.8
	.001	65	51.0
	.01	35	4.6
KI	.0001	35	8.0
	.001	30	5.6
	.01	20	2.5

The data of table 5 show also a relatively close correlation between the number of cuttings that rooted and the average total length of roots to a cutting. The data in a later part of this paper show that this correlation holds throughout the entire season where the treated cuttings are planted in the nursery.

The two reducing substances, namely $K_4Fe(CN)_6 \cdot 3H_2O$ and KI, listed in table 5 were included to test the belief of some workers that the stimulation of root growth is possibly due to the ions present in a compound and not to its properties as regards oxidation or reduction.

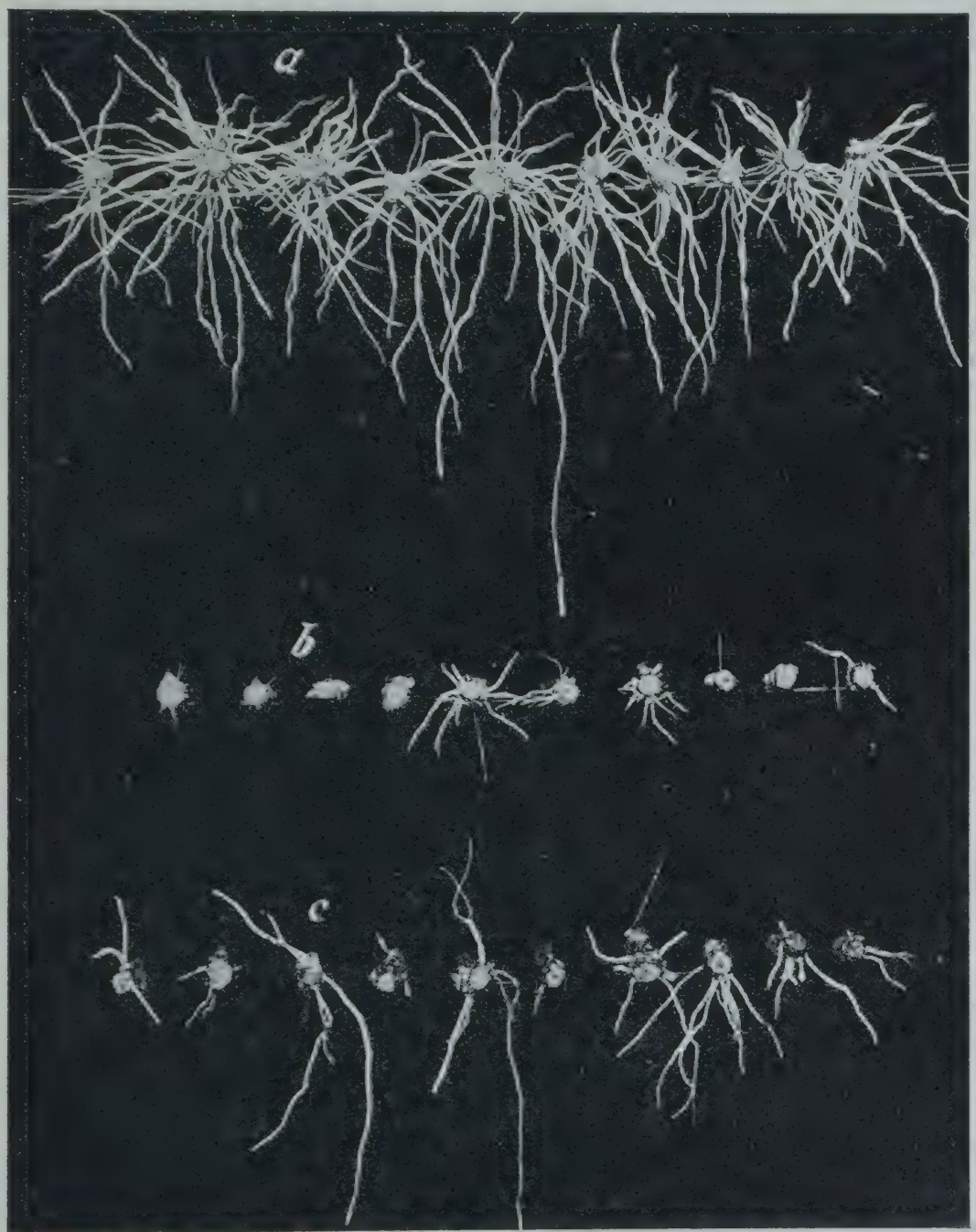

Fig. 4. The stimulating effect of $K_3Fe(CN)_6$ and $K_4Fe(CN)_6 \cdot 3H_2O$ on the development of roots. *a*, $K_3Fe(CN)_6$. *b*, Water. *c* $K_4Fe(CN)_6 \cdot 3H_2O$.

Figure 4 shows the respective stimulating effects on root development of $K_3Fe(CN)_6$ and $K_4Fe(CN)_6 \cdot 3H_2O$. The stimulation seems to be associated with the oxidizing properties of the $K_3Fe(CN)_6$, since the same ions are present in these two reagents. Similar results were obtained with the iodine and potassium iodide.

The $K_4Fe(CN)_6 \cdot 3H_2O$ did produce a slight increase in root development. This small stimulation, however, which is within the limits of experimental error, might also be the result of chemical injury.

Duration of Treatment.—Twenty cuttings were placed in an individual jar for each of the periods of treatment for every concentration

TABLE 6

THE INFLUENCE OF DURATION OF TREATMENT ON THE DEVELOPMENT OF ROOTS

Variety	Reagent	Molar concentration	Duration of treatment in hours	Per cent of cuttings with roots	Average total length of roots per cutting in centimeters after callusing
Black Monukka.....	Water	Check	24	20	2.7
(Variety of *Vitis*	Water	Check	60	20	2.5
vinifera)	Water	Check	120	10	1.0
	$KMnO_4$	.1	24	80	109.0
	$KMnO_4$	.1	60	100	143.0
	$KMnO_4$	.1	120	60	40.0
	$KMnO_4$	.25	24	60	45.0
	$KMnO_4$	.25	60	55	36.0
	$KMnO_4$	.25	120	15	31.0
	$KMnO_4$	1.0	24	30	5.0
	$KMnO_4$	1.0	60	15	1.0
	$KMnO_4$	1.0	120	0	0.0
	Water	Check	24	40	4.3
	Water	Check	48	40	4.9
	$MnSO_4$	.001	24	60	51.0
	$MnSO_4$	.001	48	100	78.0
	$MnSO_4$	.01	24	70	57.0
	$MnSO_4$	.01	48	60	45.0
	$FeCl_3$	.001	24	68	58.0
	$FeCl_3$	.001	48	80	74.0
	$FeCl_3$	.01	24	85	74.0
	$FeCl_3$	.01	48	70	67.0

of solution. At the time the cuttings of the shortest period of treatment were removed to the sand, the solutions on the others were changed. The same changes were again made when the cuttings for the next period of treatment were removed. All cuttings were callused in sand for 14 days after their removal from the solutions. The results for different periods of treatment with three different reagents are given in table 6.

The data of table 6 indicate that the duration of treatment as well as the concentration of the solution of the reagent may influence the

stimulation of root formation. That is, with a low concentration of solution, a longer period of treatment may give results comparable to a shorter treatment with a higher concentration of solution. This is well illustrated by the treatments with $MnSO_4$ and $FeCl_3$. For the 24-hour treatment, the .01 mol. solutions for these reagents gave the best results, while for the 48-hour treatment, the .001 mol. solution

TABLE 7

THE INFLUENCE OF OXIDIZING REAGENTS ON THE RATE OF ROOT AND CALLUS FORMATION ON CUTTINGS

Variety	Treatment with conc. of reagent	Treatment duration	Callusing nature and time	Per cent with roots	Average total length of roots, *cm.*	Per cent of cuttings without roots		
						Good callus	Poor callus	Without callus
Champini	Water (check)	40 hours	16 days reversed in sand pit.	0	0.0	0	40	60
	.001 m. $MnSO_4$	40 hours		100	67.0	0	0	0
	.001 m. $K_3Fe(CN)_6$	40 hours		70	87.0	20	10	0
	.01 m. Na_2O_2	40 hours		90	52.0	10	0	0
Champini	Water (check)	Continuous	20 days in the solutions in hot room.	33	12.0	15	25	25
	.0001 m. $MnSO_4$	Continuous		95	22.0	0	0	5
	.0001 m. $K_3Fe(CN)_6$	Continuous		71	49.0	15	5	9
	.001 m. Na_2O_2	Continuous		71	23.0	24	5	0
	.001 m. $KMnO_4$	Continuous		38	25.0	30	20	12
Labrusca (Pierce)	Water (check)	40 hours	18 days reversed in sand.	0	0.0	10	50	40
	.001 m. $K_3Fe(CN)_6$	40 hours		90	35.0	10	0	0
Labrusca (Pierce)	Water (check)	Continuous	20 days in the solutions in hot room.	25	5.0	25	30	20
	.0001 m. $MnSO_4$	Continuous		100	58.0	0	0	0
	.0001 m. $K_3Fe(CN)_6$	Continuous		66	12.0	15	3	10
	.001 m. Na_2O_2	Continuous		66	17.0	25	8	0
Vinifera x *Berland.* (41B)	Water (check)	40 hours	12 days reversed in sand.	0	0.0	20	10	66
	.001 m. $MnSO_4$	40 hours		20	23.0	34	25	21
	.001 m. $K_3Fe(CN)_6$	40 hours		4	20.0	34	35	33
	.01 m. Na_2O_2	40 hours		4	25.0	54	30	14
	.01 m. $KMnO_4$	40 hours		12	7.0	30	40	18

gave the greater stimulation. Similar relations of time of treatment to concentration are indicated by the tests with $KMnO_4$.

Rate of Callus and Root Formation.—In these tests only varieties were used which are difficult to root. Fifty cuttings were used in each lot. The concentration of solution and duration of treatment employed were those which gave the best results in preliminary tests. The influence of the several reagents used on the rate of callus and root formation is shown in table 7.

As shown by the figures of table 7, all of the reagents employed induced a marked stimulation in callus and root formation. In each series for each variety, the callus and root formation of the treated lots was far ahead of the checks treated with water.

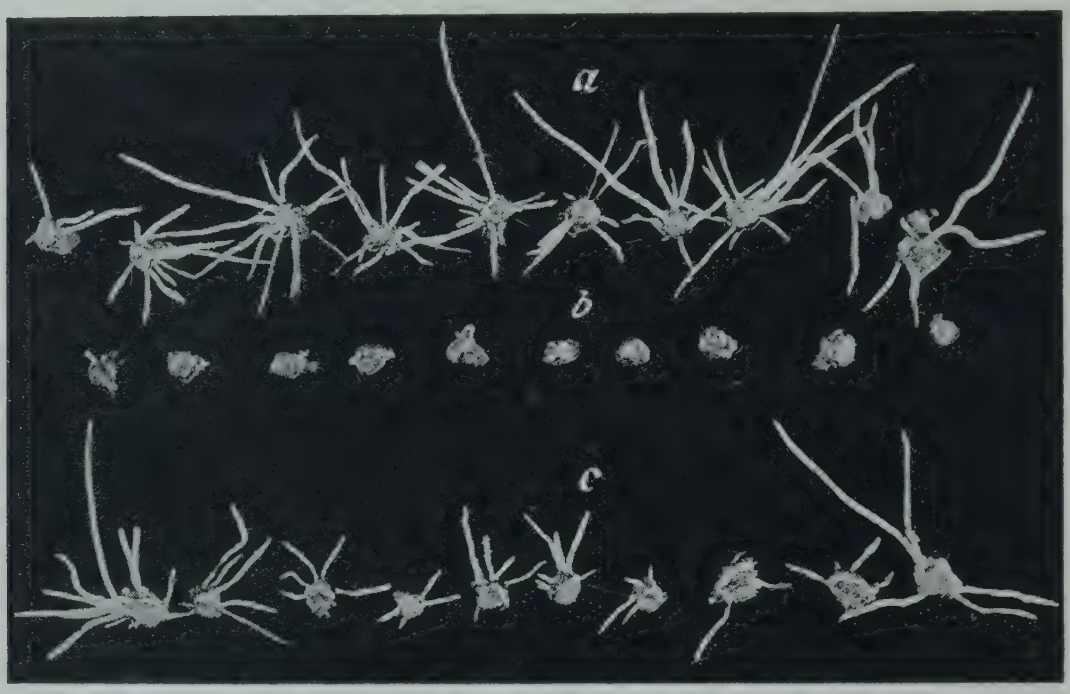

Fig. 5. The stimulating effect of $MnSO_4$ and $K_3Fe(CN)_6$ on the formation of roots on Pierce cuttings. *a*, $MnSO_4$. *b*, Water. *c*, $K_3Fe(CN)_6$.

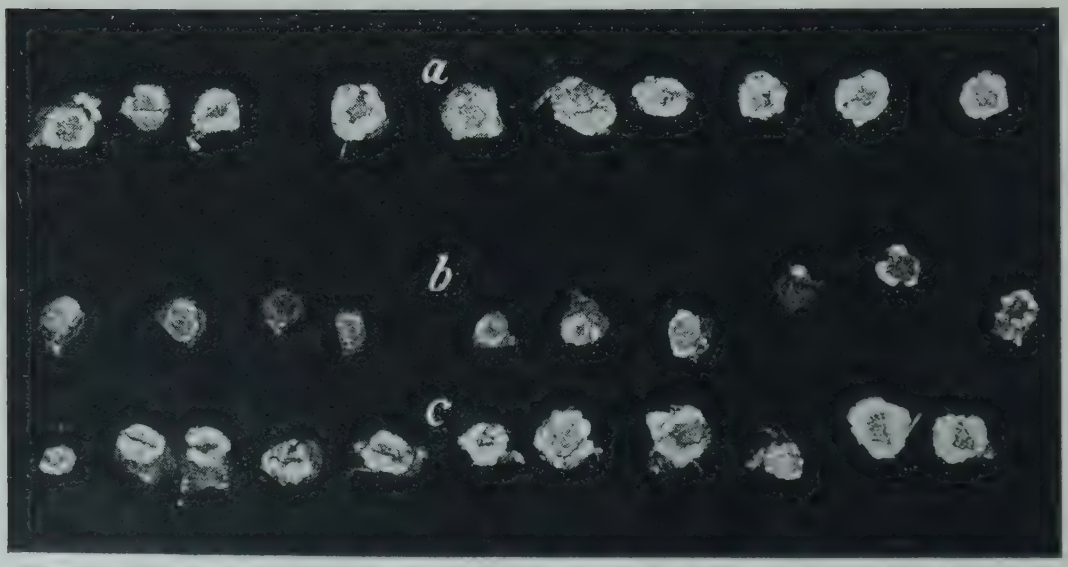

Fig. 6. The stimulating effect of $MnSO_4$ and $K_3Fe(CN)_6$ on the formation of callus on 41B cuttings. *a*, $MnSO_4$. *b*, Water. *c*, $K_3Fe(CN)_6$.

These data indicate also that in all the series in which it was employed, $MnSO_4$ gave the greatest stimulation as shown by the percentage of cuttings with roots. The $K_3Fe(CN)_6$ gave the next best stimulation of root formation, if both the percentage of cuttings with roots and the average total length of roots per cutting are considered.

The Na_2O_2 was only slighly less stimulating than the $K_3Fe(CN)_6$, while the $KMnO_4$, which has possibly received the most attention in the past as a stimulant of root growth, gave the poorest results.

The stimulating influence of $MnSO_4$ and $K_3Fe(CN)_6$ is further indicated by figure 5. These cuttings of Pierce were treated continuously for 20 days and were then held in sand 6 days before photographing. The effect of these same reagents on the formation of callus on 41B cuttings is illustrated in figure 6. Those shown here

TABLE 8

THE INFLUENCE OF CERTAIN OXIDIZING REAGENTS ON THE PERCENTAGE OF CUTTINGS THAT ROOT

Reagent	Duration of the treatment, in hours	Variety and the percentage that rooted		
		41B	Champini	Pierce
Water (check)	24	40	50	40
$KMnO_4$ .1 mol	24	100	60	.....
$KMnO_4$ .05 mol	48	90	.....	65
$MnSO_4$ .001 mol	48	100	80	100
$FeCl_3$ .01 mol	24	80	60	.....
$K_3Fe(CN)_6$ .001 mol	24	100	87	80
Sand (planted direct from sand pit)	.....	25	30	20

are ten average cuttings of the lots listed in table 7 for these treatments of this variety. The photograph, however, was taken after the cuttings had been in the sand for only 8 days, which was before any roots had formed. It is, of course, a well known fact that a heavy formation of callus does not necessarily indicate that root formation will follow, or that the absence of callus precludes the formation of roots. Rooting may be independent of callus formation. However, as is shown by figures 7 and 10, the formation of callus on these cuttings was an indication of their respective rates of development.

Number and Vigor of the Rootings Produced.—Fifty cuttings of each variety were used for each of the reagents. The cuttings were treated 40 hours in the solutions and then callused in sand before planting. In the nursery the cuttings were planted six inches apart in rows spaced six feet. The influence of the treatment on the number and vigor of the rootings for the varieties tested during 1924 is given in tables 8 and 9.

The data of table 8 show a considerable increase in the percentage of cuttings that rooted for each of the varieties as a result of the treatments. The greatest increases were obtained in 41B and the least in Champini. There was little difference in the stimulating effect of the several reagents.

Of equally as great significance as the increase in the percentage of rootings produced is the increase in the number of first class or vigorous rootings. Some measurements made at the end of the first

TABLE 9

THE INFLUENCE OF CERTAIN OXIDIZING REAGENTS ON THE VIGOR OF THE ROOTINGS

Variety	Treatment	Per cent of rootings that were vigorous	Relative circumference increase in centimeters	Total length of top growth in centimeters	Average diameter of individual roots in mm.
Champini	Check (in water).....	28	.4	166	.....
	$MnSO_4$ .001 mol.....	88	2.4	439	.....
	$FeCl_3$ .001 mol.....	83	2.3	490	.....
	$KMnO_4$ .05 mol.....	83	1.5	416	.....
	Check (planted directly from sand pit).....	0	.7	122	.....
41B.....	Check (in water).....	20	1.3	.....	1.9
	$MnSO_4$ .001 mol.....	80	2.5	.....	3.6
	$FeCl_3$ .01 mol.....	75	2.7	.....	3.3
	$KMnO_4$ .05 mol.....	75	2.7	.....	2.9
	Check (planted directly from sand pit).....	5	.3	.....	1.4

season's growth in the nursery on the difference in the vigor of the rootings produced by the treated lots as compared to the checks are given in table 9.

The data of tables 8 and 9 indicate that the increase in vigor was more pronounced than the increase in the percentage of rootings. That is, 41B gave a rooting percentage of 40 in water and 100 in .001 mol. $MnSO_4$ (table 8), while the percentage of vigorous rootings was 20 and 80, respectively (table 9). Similarly, the Champini in water had a rooting percentage of 50 and in $MnSO_4$ 80, while the percentage of vigorous rootings produced under these treatments was 28 and 88, respectively. The same is true for the other treatments.

The influence of the several oxidizing agents employed, on the number and vigor of the rootings produced during 1925, was similar to that of 1924. Additional data, however, were collected and

the probable error of some of the measurements determined; hence, the results may serve as a better basis of comparisons than those of tables 8 and 9. The influence of these treatments on the number and vigor of the rootings produced is given in table 10 and the influence on root develepment is illustrated further in figures 7 and 10.

TABLE 10

THE INFLUENCE OF CERTAIN OXIDIZING REAGENTS ON THE NUMBER AND VIGOR OF ROOTINGS PRODUCED

Variety	Treatment	Mol. conc. of reagents	Per cent of cuttings rooted	Per cent of rootings that were vigorous	Number of good rootings from 100 cuttings	Circum. increase in centimeters	Average total length of top growth *cm.*	Average number of roots to a plant	Average diameter of individual roots *mm.*
Champini	Water		70	50	35	.53±.14	140±24	5.8	2.8±.23
	$MnSO_4$	.001	100	100	100	1.11±.16	244±13	7.3	3.8±.20
	$K_3Fe(CN)_6$	.001	100	90	90	1.37±.21	312±24	8.8	4.1±.18
	Na_2O_2	.01	100	100	100	.97±.17	313±17	7.8	3.8±.20
	$KMnO_4$	.01	85	90	76	.85±.13	230±15	7.1	3.5±.16
Pierce	Water		52	40	21	.39±.07	71±8.4	6.4	2.3±.09
	$MnSO_4$	.001	100	90	90	.86±.11	204±16	6.8	3.1±.17
	$K_3Fe(CN)_6$	.001	100	87	87	.72±.10	185±10.2	7.7	2.8±.18
	Na_2O_2	.01	75	66	50	.65±.08	155±13	5.7	3.6±.22
	$KMnO_4$	.01	65	50	33	.54±.08	142±8	6.2	2.9±.15
41B	Water		30	10	3	.67±.07	47±10.2	2.7	2.0±.19
	$MnSO_4$	.001	95	85	80	1.09±.10	218±33	5.6	3.3±.21
	$K_3Fe(CN)_6$	.001	90	89	80	.97±.11	192±15	6.4	3.3±.19
	Na_2O_2	.01	75	70	52	.93±.13	196±17	5.7	3.1±.13
	$KMnO_4$	.01	65	72	47	.86±.07	144±16	4.8	2.8±.17

The data of table 10 show very marked increases in both the number and the vigor of the rooting produced in the treated lots as compared to the checks. Here again there was little difference in the results obtained for the different compounds, except that the results with $KMnO_4$ were somewhat less marked than those for the other three reagents.

The matter of greatest importance in the possible application of these treatments to practice, however, is the increase in the number of vigorous rootings produced by each 100 cuttings planted. It has been shown by Bioletti(1) that the strongest 25 per cent of 630 Muscat rootings produced 50 per cent more crop at the first vintage than the weakest 25 per cent of these rootings. He believes that the advantage of the strongest rootings was in reaching nearly full bearing the third season instead of the fourth as with the weaker rootings.

The increases over the check in Champini in the number of vigorous rootings were 186, 171, 186, and 117 per cent, respectively, for the $MnSO_4$, $K_3Fe(CN)_6$, and Na_2O_2, and $KMnO_4$. In 41B and Pierce, the increases for the treatments used were even greater.

The increases in circumference of the cuttings treated with $MnSO_4$, $K_3Fe(CN)_6$, Na_2O_2, and $KMnO_4$ exceeded those of the check in Champini by 109, 158, 83, and 68 per cent, and in Pierce by 112, 82, 67, and 38 per cent, respectively. Similar increases were realized with 41B.

Fig. 7. The influence of $MnSo_4$ and $K_3Fe(CN)_6$ on the number and vigor of the roots produced by Champini cuttings at the end of the growing season. *a*, $K_3Fe(CN)_6$. *b*, Water. *c*, $MnSO_4$.

The average total length of top growth to a rooting exceeded that of the check in Champini by 74, 123, 124, and 64 per cent; in Pierce by 187, 161, 118, and 100 per cent; and in 41B by 356, 308, 317, and 206 per cent, respectively, for the treatments with $MnSO_4$, $K_3Fe(CN)_6$, Na_2O_2, and $KMnO_4$.

Similar increases were obtained in the number and average diameter of the roots. Although the check cuttings produced the smallest number of roots in all but two instances, these roots in no case had as great an average diameter as the roots of the treated lots.

The greater vigor and possibly the greater number of rootings produced are not the result only of a stimulated growth in the commencement of root formation, but also of a more rapid vegetative growth, which is apparent throughout the development in the nursery. This greater vigor of growth during early summer in the nursery is shown by figures 8 and 9. Figure 8 shows the relative development

of the cuttings shown in figure 6 five weeks after planting. The differences in top growth were equal to, if not greater than, the differences in callus formation as illustrated in the former figure. Figure 9 shows the top development of 41B cuttings eight weeks after plant-

Fig. 8. Showing the influence of treatment with $MnSO_4$ and $K_3Fe(CN)_6$ on the rate of growth of 41B cuttings in the nursery during early summer. Five weeks after planting. *a*, $MnSO_4$. *b*, Water. *c*, $K_3Fe(CN)_6$.

Fig. 9. The influence of treatment with $MnSO_4$ and $KMnO_4$ on the rate of growth of 41B cuttings in the nursery during early summer. Eight weeks after planting. *a*, $MnSO_4$. *b*, Water. *c*, $KMnO_4$.

ing. Here again the more rapid development, in the nursery, of the treated cuttings is pronounced.

The development of the cuttings of 41B shown in figures 6 and 8 at the end of the growing season is illustrated by the roots shown in figure 10. As this figure indicates, the more rapid development of the treated cuttings appears to continue throughout the growing season.

An Apparent Difference in the Stimulating Action of Different Oxidizing Reagents.—During some of the tests in continuous treatments, cuttings were immersed as much as four inches in the solutions of the reagents. It was observed that under these conditions the stimulation of root growth at the base of the cuttings by the reagents whose formulae show oxygen was greater than that by those whose formulae do not.

TABLE 11

THE STIMULATION OF ROOT GROWTH BY OXIDIZING REAGENTS IN THE PRESENCE OF AIR

Reagent	Mol. conc.	Average number of roots per cutting	Average total length of roots per cutting, centimeters
$MnSO_4$	.001	3.0	43
H_2O_2	.01	2.5	33
$FeCl_3$	.01	2.5	47
$K_3Fe(CN)_6$	.01	3.0	37

TABLE 12

THE EFFECT OF IMMERSION ON THE STIMULATION OF ROOT FORMATION BY OXIDIZING REAGENTS

Reagent	Percentage of cuttings with roots at base	Percentage of cuttings with roots at surface of the liquid	Percentage of roots formed at base of the cutting
Check (water)	10	100	5
$MnSO_4$ .0001 per cent	90	30	95
H_2O_2 .0001 per cent	80	0	100
$K_3Fe(CN)_6$ .0015 per cent	50	80	25
$FeCl_3$ .001 per cent	20	80	10

In the presence of air, the oxidation of the tissue of the cuttings appears to be independent of the presence or lack of oxygen in the formulae of the reagents. For instance, when cuttings were treated and then callused or rooted in sand, $K_3Fe(CN)_6$ and $FeCl_3$ stimulated root growth as much as $MnSO_4$ or H_2O_2. This is illustrated by the number and vigor of the roots produced as indicated in table 11.

On the other hand, when cuttings were rooted in the absence of air, as in water or in the solutions of a reagent, a considerable difference in the stimulation of the root growth became apparent between the reagents whose formulae show oxygen as compared to those whose formulae do not. This difference is indicated by the photo-

Fig. 10. The influence of treatment with $MnSO_4$ and $K_3Fe(CN)_6$ on the development of roots on 41B cuttings at the end of the growing season. *a*, $MnSO_4$. *b*, Water. *c*, $K_3Fe(CN)_6$.

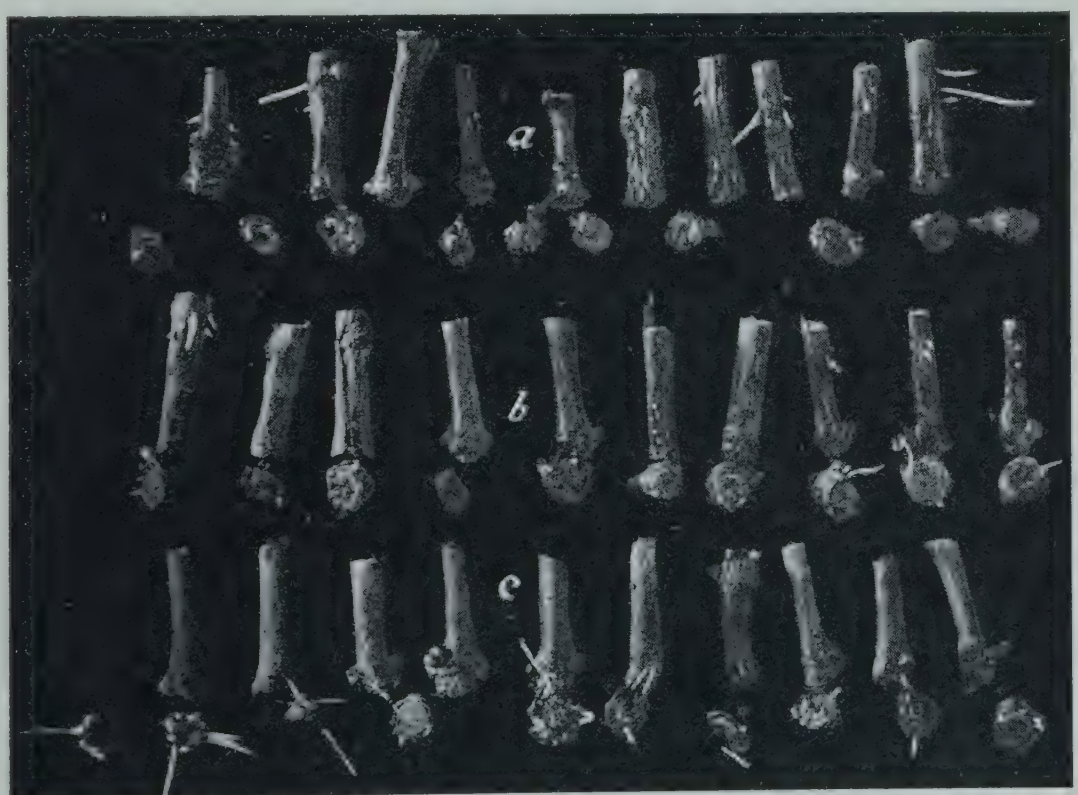

Fig. 11. The influence of the presence or lack of oxygen in the formulae of the reagents on the position of root formation on Pierce cuttings when rooted in the solutions of the reagents. Cuttings immersed one and one-half inches into the solutions. *a*, Water. *b*, $K_3Fe(CN)_6$. *c*, $MnSO_4$.

graphs of figure 11 and the data of table 12, which were obtained by immersing Pierce cuttings one and a half inches in water and solutions of the reagents, and by holding them at 30° C until roots developed. The solutions were changed every 48 hours.

The data of table 12 indicate that the stimulation of growth by the reagents not showing oxygen in their formulae is greatly retarded by the absence of air. In the case of the reagents carrying oxygen, however, there is a very decided stimulation in root growth even in the absence of air. As indicated by Rosa(3) there seems to be a specific effect of the oxygen, aside from the liberation of positive charges through the reduction of a salt or ion absorbed by the tissues of the cutting. That is, the reagents showing oxygen in their formulae seem to be capable of a certain amount of oxygenation of the tissues in addition to oxidation in the strict meaning of the term.

SUMMARY

1. Selection by means of the iodine-starch test has resulted in a marked increase in the percentage of cuttings that root. The number of rootings of the cuttings showing a relatively high starch content was 264 per cent of that of the cuttings low in starch.

2. Chemical analyses indicate that the iodine-starch test gives a relatively accurate indication of the starch stored in the cuttings.

3. Early planting resulted in an increase in the number and vigor of vine rootings. The early planting appears to improve the development of the cuttings through its effect on the time of beginning of root formation.

4. The greatest stimulation of root development by the oxidizing reagents with a 24-hour treatment was obtained with the following range of concentrations: .001 to .0001 mol. $MnSO_4$, $Mn_2(SO_4)_3$, $K_3Fe(CN)_6$, and iodine; .01 to .001 mol. MnO_2, $FeCl_3$, and Na_2O_2, and .1 to .05 mol. H_2O_2 and $KMnO_4$.

5. The time required for treatment, within certain limits, is a function of the concentration of the solutions of the reagents.

6. Oxidizing reagents hastened both callus and root formation.

7. Oxidizing reagents improved the rooting of cuttings that root with difficulty.

8. The number of vigorous rootings of Champini was increased 186, 171, 186, and 117 per cent, respectively, by treatment with $MnSO_4$, $K_3Fe(CN)_6$, Na_2O_2, and $KMnO_4$. In 41B the increases were even greater.

9. Treatment with oxidizing reagents resulted in marked increases in the relative circumference of the cuttings and the average total length of top growth at the end of the first season in the nursery.

10. The development of the treated cuttings appears to be more vigorous than that of the untreated throughout the growing season in the nursery.

11. There appears to be a greater stimulation of root growth at the base of cuttings by the reagents whose formulae show oxygen than by those whose formulae do not, when the cuttings are rooted in the solutions of the reagents.

LITERATURE CITED

(1) Bioletti, F. T.
1926. Selection of planting stock for vineyards. Hilgardia 2:1–23.

(2) Curtis, O. F.
1918. Stimulation of root growth in cuttings by treatment with chemical compounds. Cornell Univ. Agr. Exp. Sta. Memoir **14**:69–138.

(3) Rosa, J. T.
1923. Abbreviation of the dormant period in potato tubers. Proc. Amer. Soc. Hort. Sci. **20**:180–187.

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A PRELIMINARY STUDY OF PETROLEUM OIL AS AN INSECTICIDE FOR CITRUS TREES[1]

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INTRODUCTION

The general purpose of this investigation was a study of petroleum oils in relation to their availability as insecticides for use on citrus trees. This involved selection, first on the basis of tree tolerance, and secondly on the basis of insecticidal value. The data thus far obtained and the conclusions derived therefrom are believed to be of sufficient importance to justify this preliminary report. Investigations along the more promising lines opened up by the study are still in progress.

While the several phases of this investigation were conducted coöperatively and with free consultation between the authors, portions of the work were of necessity carried out semi-independently. Thus the selection of oils by foliage testing and the development of the chemical aspect of the paper have been largely the work of deOng at Berkeley and in southern California, while the insecticidal tests proper, together with their accompanying developments, were principally the joint work of Knight and Chamberlin at Riverside. The authors are indebted to H. J. Quayle for valuable suggestions and criticisms.

The study of petroleum-oil distillates in relation to their insecticidal effects was originally begun at the California Agricultural Experiment

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Station in 1914 but was later discontinued until 1924 because of lack of facilities. At that time a coöperative project was undertaken between the University of California Agricultural Experiment Station and the Standard Oil Company of California. Under this coöperative plan, the Standard Oil Company contributed the services of their oil chemists and the use of their laboratories in preparing oil samples for both laboratory and field tests. The Experiment Station was responsible for testing the various oils with reference to insect kill and plant tolerance.

A special advantage that has been realized from thus coöperating with a large commercial company lies in the fact that the distillates and oils were obtained from crude oils pooled from a large number of wells. Hence the samples tested may be safely considered representative of supplies which might be secured from any large oil company of California. This overcomes one of the principal difficulties in the study of petroleum oils, viz., that the oils used were not necessarily typical of supplies generally available.

HISTORY OF OIL SPRAYS IN RELATION TO CITRUS TREES

The use of petroleum-oil sprays as insecticides on citrus trees began about 1881 when kerosene emulsions first came into use. This type of oil has been found safe to use on citrus trees but does not control the more resistant scale insects and mealybugs. In an attempt to find a cheaper and more toxic oil, use was made of the so-called "stove distillates," these being unrefined distillates of 26° to 32° A.P.I.[5]

Severe injury to the fruit as well as to the foliage was frequently caused by these materials when emulsified with soap, as was ordinarily done, and when used in the form of a mechanical mixture of oil and water. Such mechanical mixtures of oil and water are, as their name implies, formed by violent agitation in the spray tank without the use of emulsifying agents. This type of "emulsion" separates almost instantaneously and it was found in practice that pure oil would occasionally be applied to parts of a tree with severe injury resulting. For some years, therefore, the spraying of citrus trees for the control of insects was of doubtful value. The perfection of fumigation with hydrocyanic acid gas also tended to discourage the use of other scalecides. The high cost of fumigation has, however, from its inception led to sporadic attempts to find an effective substitute.

[5] A.P.I.—American Petroleum Institute.

The factor which has probably done most to stimulate recent investigations of sprays has been the development of HCN-resistant strains of both the red and the black scale insects, as shown by Quayle.(1) These resistant strains show a distinct tolerance to dosages of hydrocyanic acid gas which in earlier fumigation practice were found to be fatal. Thus the scale kill by fumigation in the areas of resistance has dropped from over 99 per cent to as low as 85–95 per cent, so that the control of 85–96 per cent now commonly attained by spraying is hopeful, although below the standard of successful fumigation.

Gray(2) in 1915 noted the important relation existing between the refining of petroleum oil by the use of sulfuric acid and plant tolerance as discussed further on in this paper.[5a] The cost of highly refined white oils, however, prevented their effective utilization in the stable emulsions of high oil content. The development of a successful quick-breaking emulsion, as shown later, affords an effective means of overcoming this objection by reducing the necessary concentration of oil, and by permitting its more complete utilization.

COMPOSITION OF PETROLEUM OILS AND THEIR INJURY TO PLANTS

Nature of Injury to Trees by Petroleum Oils.—Volck(3) has shown that injury is most pronounced when the application of oil is made to the under side of the orange leaf where all the stomata of this plant are situated. This is owing to the fact that oil penetration into a leaf is much facilitated by any sort of opening, abrasion, or pore. According to the work of Magness and Burroughs(4) an oil film on the surface of stored apples may have a distinct effect on the gaseous exchange. The evolution of carbon dioxide from Winesap apples held at 65° F was reduced only 12 per cent by a coating of Oronite Crystal or other petroleum oil, but analyses of the air in the intercellular spaces showed a composition of 2.6 per cent oxygen and 25.3 per cent carbon dioxide, while check apples had 5.7 per cent oxygen and 18.3 per cent carbon dioxide. Burroughs(5) has noted a reduction in the amount of starch produced in apple leaves that seem to have been arrested in their growth by the application of an oil spray.

Gray and deOng(2) found a correlation between the specific gravity of the oil and resulting foliage injury. This correlation

[5a] A commercial emulsion of highly refined petroleum oil was being made by W. H. Volck when this later investigation by the California Experiment Station was begun in 1924.

applies only in a comparison of kerosenes with the heavier or lubricating oils. The former have a much lower boiling point and volatilize before penetration occurs; or even if penetration does take place, the oil may still volatilize before injury results. Injury may be possible, however, with certain fractions of a still lower boiling point than kerosene, especially if they contain a high percentage of unsaturated hydrocarbons. A study of lubricating oils having a much higher range of boiling points than kerosenes shows that their effect on plants and insects is more nearly related to viscosity than to specific gravity.

Injury to the foliage of citrus trees from petroleum oils is of two distinct types, acute and chronic. The former is caused by light (low-boiling-point) oils, the latter by heavy (high-boiling-point) oils. In the acute type of injury two distinct phases are noticeable. First, the leaf tissue may be killed within 48 hours after the application; and secondly, this may be followed by the leaves dropping after three or four days, although such leaves do not lose their color to any marked degree. Injury to the fruit or wood seldom occurs except with oils having a high percentage of unsaturated hydrocarbons, such as is commonly found in untreated oils, or those slightly refined. Contact of these oils with the roots may cause the death of the tree within a relatively short time.

Chronic injury is associated in varying degrees with oils of a high boiling point, which leave an oil film on the leaf and twig surface for a period of days or weeks. The foliage becomes yellow and defoliation begins within a few days and may last for weeks. The twigs and even the larger limbs are stunted or killed as shown in figure 1. The orange tree in the figure was photographed one year after a portion of it had been sprayed with lubricating distillate, untreated with sulfuric acid.

The damage occurred only on the sprayed portion. The normal growth in the background, which shows no sign of injury, was unsprayed. Stunted twigs often put out a few small, weak leaves, and frequently the tree sprouts freely just below the injured parts.

Fully mature leaves, especially if senile, are more susceptible to injury from neutral oils than younger ones which are still in the growing stage. On the other hand, very young leaves are much more susceptible to injury from unrefined oils than the mature, but not senile, leaves, although this varies a great deal according to the degree of refinement of the oil used.

Commercial Refining of Oils as a Means of Reducing Injury to Plants.—Petroleum distillates are not usually in a marketable condi-

tion without chemical treatment to remove such ingredients as sulfur, resinous matter and the unsaturated and aromatic hydrocarbons. The common refinery practice is to treat the raw distillates, resulting from the heating of crude oil, with sulfuric acid. The quantity of acid

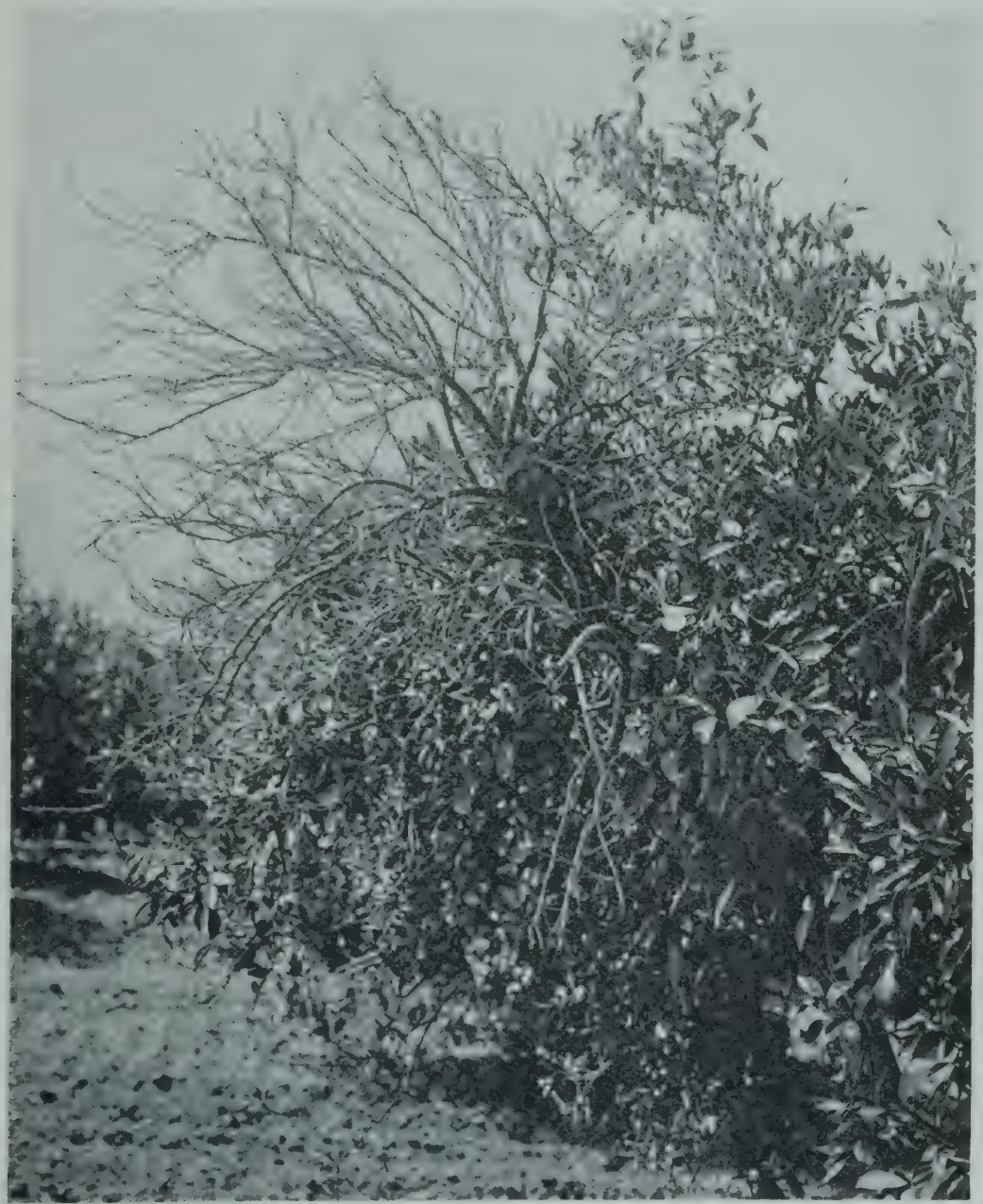

Fig. 1. Orange tree showing dead wood where the tree was sprayed with unrefined oil distillate.

required and the length of time during which treatment is continued depend on the grade of product desired and on the purity of the distillate used.

After treatment with acid, the oils are washed with a solution of caustic soda in order to remove the unchanged petroleum acids and phenols and to neutralize and remove the sulfo-acids and the sulfuric

acid remaining in the oil. The expense of refining operations is high, especially for the highly refined white oils, since the acid sludge resulting from the treatment is largely a waste product. In addition to the cost of the acid there is the loss of distillate which may amount to more than 50 per cent of the original volume treated.

In order to avoid the losses resulting from the chemical treatment, it is advisable to substitute a method of extraction more nearly of a physical type. This has been accomplished by Edeleanu(6) who found that if petroleum oil was treated with liquid sulfur dioxide the olefins are dissolved but the saturated hydrocarbons remain unaffected. Experimental work is now in progress with oils refined by this method in the hope that a satisfactory degree of refinement may be obtained at a lower cost than is possible from the sulfuric acid treatment.

Tests of Petroleum Oils in Relation to Plant Injury.—Differences in the insecticidal effects of, and plant tolerance for, various petroleum distillates have long been recognized, but until recently the only specifications commonly used for distinguishing between them were specific gravity and, perhaps, the flash point. We now know that these are inadequate criteria. The boiling point of different kerosenes, for example, has been shown by Moore(7) to be important in distinguishing between the toxicity of petroleum fractions as it relates both to insects and to plants.

Since some progress has been made in determining the relation between the unsaturated hydrocarbon content of oils and their toxicity to plants, our first attempt at selection was based on the degree of refinement, the assumption being that the oil fractions containing the least amount of sulfonatable oil would be the safest.

A series of lubricating and kerosene oils was obtained from the Standard Oil Company. These ranged from the raw untreated distillate resulting from the distillation of crude oil, up through the different degrees of refinement effected by the use of acid and filtration. Their physical and chemical specifications are shown in table 1.

These oils, with the exception of the one finally selected for study, are referred to by number throughout this paper, instead of by their trade names. Oils 1 to 6, inclusive (tables 1 and 2), are lubricating oils, oil 1 being the raw distillate, and oil 5 the end of the refined series as based on the sulfonation test. Oil 5 (Oronite Crystal) is a very bland and "neutral" oil; it is colorless, odorless and tasteless. Oils 1a to 4a are kerosenes arranged in the same way, oil 1a being the raw distillate and 4a the highly refined end product. The sulfonation test (table 2) shows the amount of unsaturated hydrocarbons present in the various samples. These oils were emulsified with

TABLE 1

PROPERTIES[a] OF OILS TESTED

No.	Gravity[b] (degrees A. P. I.)	Flash point[c] ° F	Fire point ° F	Viscosity[d] in seconds at 100° F	Color	Sulfur (per cent)	Unsulfonated residue of oil	Acidity in mg. of KOH per gm. of oil
1	19.2	305	.....	105	.....	.7	51	1.5
2	21.3	310	350	99	.....	.65	52	1.0
3	22.5	310	350	96	6[e]	.6	56	.6
4	22.7	320	360	107	2.5−	.6	60	.4
4x	22.7	320	360	100–110	1.5	.6	62	.03
5[h]	29.8	320	360	106	+25[f]	.006	98	.0
5x[h]	28–31	280+	.....	70–80	+25[f]	.015	98	.0
6	.....	360+	.....	330–340	3.	.6	58	.2
KEROSENES								
1a	35.9	124CT[g]	.....	375	.....	.....	81	.....
2a	41	83CT	125	320	+25[f]	.016	82	.....
3a	41.3	113CT	135	345	+25[f]	.010	93	.....
4a	43.3	143CT	175	400	+25[f]	.006	98	.....

[a] The pour point on all oils used was below zero Fahrenheit.

[b] The A.P.I. gravity table is so similar to the Baumé gravity table that for all practical purposes they may be considered identical for lubricating oils.

[c] Cleveland open cup.

[d] Viscosity of lubricating oils determined by the Saybolt Universal viscosimeter. Viscosity of kerosene fractions measured by the Saybolt "Thermoviscosimeter," which bears no relation to the lubricating-oil viscosimeter.

[e] A.S.T.M. standards.

[f] A.S.T.M. standard for kerosene by Saybolt colorimeter; the color number + 25 is an arbitrary value given to the most highly refined kerosenes.

[g] Closed Tagliabue Tester.

[h] No. 5 is Oronite Crystal oil, No. 5x, Oronite Cosmetic oil, trade names used by the Standard Oil Company of California to designate oils of the above specifications. Other numbers used in the table also refer to commercial brands of oil as sold by the Standard Oil Company. Note the gravity of Oronite Crystal oil (No. 5). This increase (from about 22.5° A.P.I. to 29.8°) is the result of the excessive treatment with acid. It follows that gravity alone is no criterion since this property of finished white oil is practically identical with that of some raw distillates such as gas oil, though obviously viscosity and flash (and molecular weight) are much higher. As a general rule the saturated hydrocarbons are much lighter in gravity than either olefins or aromatics, e.g., heptane (N), gravity 75° and toluene, gravity 32° A.P.I.

sodium oleate and applied to the trees with a hand sprayer at an oil concentration of 6 per cent. Field tests on orange and lemon trees were made during the season of 1924 at Riverside, California, where the typical high temperatures and low humidities of southern interior California occur; at Lindsay as typical of the upper San Joaquin Valley; and also at Santa Paula, which has the lower temperature characteristic of coastal conditions. The maximum temperature at the last point usually ranges 10° to 15° F lower than at Riverside. The latter experiments were made possible by the coöperation of Mr. C. T. Dodds of the Santa Paula Citrus Fruit Association. The results are clearly brought out in table 2.

TABLE 2

EFFECT OF PETROLEUM OILS OF DIFFERENT VISCOSITIES AND SULFONATION VALUES ON ORANGE TREES AT VARYING TEMPERATURES.*
OIL CONCENTRATION IN FISH-OIL SOAP EMULSION SIX PER CENT†

Oil No.	Viscosity in seconds at 100° F	Sulfonation value,‡ per cent	Riverside				Santa Paula	
			Series I		Series II		Series I	
			Max. temp. range 85–100° F Applied 6/29		Max. temp. range 87–104° F Applied 8/21	Max. temp. range 61–96° F Applied 10/31	Max. temp. range 80–90° F Applied 6/16	
			Observations made 7/9	Observations made 8/2	Observations made 8/30	Observations made 11/9	Observations made 6/23	Observations made 7/21
1	105	51	Severe defoliation and burn.	70% twigs dead, 20% defoliated but sprouting.			12% defoliation, 40% fruit scarred.	10% fruit scarred, 25% leaf burn.
2	99	52	Severe defoliation and burn on twigs and fruit.	60% twigs dead, 20% defoliated but sprouting.			8% defoliation	40% fruit scarred, 20% leaf burn.
3	96	56	Slight defoliation, no burn.	5% twigs dead, 10% defoliation.			2% defoliation	20% fruit scarred, 15% leaf burn.
4	107	60	Heavy defoliation, no burn.	10% twigs and fruit dead, sprouting next injured area.	15% defoliation, no twig or fruit injury.		12% defoliation, 30% fruit scarred.	No fruit scarred, 8% leaf burn.
4x	105	62	Same as No. 4	Same as No. 4	12% defoliation, no twig or fruit injury.		15% defoliation, 40% fruit scarred.	15% leaf burn, 20% fruit scarred.
5	106	98	Slight defoliation, but showing growth.	Normal	2% defoliation		1% defoliation	Normal.
5x	75	98	Slight defoliation and twig burn, no fruit injury, new growth normal.		5% defoliation		3% defoliation	Almost normal.
6	330	58	Severe defoliation and burn, no new growth.	35% twigs dead, defoliated area sprouting.	12% defoliation	Normal	5% defoliation, 10% fruit scarred.	10% fruit scarred, 12% leaf burn.
1a	375	81	Normal	3% defoliation			Normal	Normal.
2a	320	82	Normal	Normal			Normal	Normal.
3a	400	93	Normal	Normal			Normal	Normal.
4a	400	98	Normal	Normal			Normal	Normal.

*Defoliation values are estimates only and are given in percentages for convenience in recording. † This is a stable emulsion. ‡ Amount of oil soluble in 37N H_2SO_4.

On the basis of these experiments it was possible to eliminate all those lubricating oils which did not show a high degree of refinement. The data in table 2 show that the first four oils used were dangerous to fruit and foliage and even to the tree itself. As expected, the degree of injury corresponded very closely to the amount of sulfonatable oil present. Filtration through Fuller's earth seemed to have no effect whatever in reducing injury. For example, oil 4x is a filtered oil of the type of oil 4 and, although variation in the degree of injury resulting from the use of these two oils is sometimes seen, a comparison of the effects produced in all field work thus far shows that no distinction can be drawn between them. Oils 5 and 5x are very similar from the sulfonation standpoint, but the latter is less viscous and has a lower boiling point. It "evaporates" or rather disappears from the foliage more quickly than the former and for that reason is possibly the safer. This disappearance of an oil film from foliage is not a simple phenomenon. It seems probable that it is due primarily to absorption followed by oxidation rather than to simple volatility. This point requires much further investigation before any safe generalization can be drawn. Oil 6 has a high viscosity and boiling point and is not very highly refined, and thus caused serious injury, especially at high temperatures.

These data show that only the most highly refined lubricating oils (such as 5 and 5x) are safe enough to justify experimentation on citrus trees at summer temperatures.

Under summer conditions at Riverside, the leaf drop may begin from a week to ten days after spraying. Under coastal conditions (i.e., at Santa Paula) it may be delayed six weeks or more. Under winter temperatures with maxima of 50° to 70° F, oils of lower refinement and higher boiling point are safe to use. As a result of these tests our succeeding work involved primarily a close study of oil 5, known commercially as Oronite Crystal oil.

The kerosene type of oil shows a reaction similar to the lubricating oil, in that the raw distillate 1a was more injurious than any of the three oils 2a to 4a, having various degrees of refinement. It will be noticed that 2a is less injurious than 3a, probably owing to its greater volatility. These kerosenes are all very much safer to use than the lubricating oils, but on account of their low boiling point they evaporate relatively quickly and hence are not satisfactory scalecides except possibly for the very youngest stages of scale insects.

In general, these tests indicate that a petroleum lubricating oil, to be safely used on citrus foliage in summer, must be of a very high degree of refinement and neutrality. The "white" lubricating

oils such as Oronite Crystal and Oronite Cosmetic most nearly meet this requirement. It is also evident that most kerosenes are safe under ordinary conditions but these oils must be eliminated because their volatility limits their insecticidal value. Their lack of injury to foliage is also in large part to be ascribed to their volatility.

PHYSIOLOGICAL EFFECT OF NEUTRAL WHITE OILS ON CITRUS TREES

While the neutral white oils such as Oronite Crystal have been spoken of as non-toxic, nevertheless, their presence upon a citrus tree sometimes induces certain characteristic effects which are more or less deleterious. These effects have not been studied enough to be at all adequately understood, and hence the following statement is mainly descriptive.

The most characteristic effect is a more or less heavy leaf drop, principally of senile or semi-senile leaves. For the most part this seems to be an acceleration of a normal process. It occurs on both oranges and lemons.

The next most characteristic effect consists of fruit "injury," particularly to lemons. The most common effect is the dropping of tree-ripe fruits, which is analogous to the dropping of senile leaves. A second and more important kind of fruit injury is a more or less marked delay in the coloring of green lemons subjected to the ethylene gas treatment. In some instances this delay is almost or quite permanent. This has not yet been satisfactorily explained, but it is apparently correlated with a morphological change in the oil cells in the rind of the fruit. The effect seems to consist in a withdrawal of the essential oil contained in the oil cells and may be due to its extraction by the spray oil. As shown by Fawcett(8) in 1916 the application of its own essential oil to the rind of a growing lemon inhibits or entirely prevents normal coloration.

In certain coastal areas, notably in Orange County, it is now well known that a drop of green Valencia oranges may follow application of the Oronite Crystal oil, particularly during humid weather conditions. Furthermore, ripening may be considerably retarded.

Various other pathological phenomena are continually being ascribed to the use of this oil on citrus trees, in addition to the well established ones given above. These include claims of such effects as reduction in set of fruit, actual twig, leaf, and fruit burn, dropping of newly set fruit (analogous to ordinary heat-induced "June drop") and so on, but the data available at present are too contradictory to be evaluated without further study.

THEORY OF OIL EMULSIONS

Petroleum oil is an insecticide of great value, but on account of its inherent danger to the plant, when used in effective amounts, it has been found necessary to dilute it with a material which acts as a carrier. Water lends itself readily to this purpose, but since these two liquids are immiscible it is necessary to employ some chemical or mechanical means of dispersing the oil in droplets uniformly throughout the water.

There are two types of oil emulsions. In the first, the "oil-in-water" type, the oil is dispersed as small globules throughout the water. In the second, the "water-in-oil" type, the reverse system is found. The prevailing type of oil emulsion used in insecticidal work is of the first or "oil-in-water" type, the invert form having been studied only very recently.

The nature of the emulsion, whether of the ordinary or invert type, is determined by the kind of emulsifier as has been shown by Tinkle, Draper, and Hildebrand(9) and Bhatnagar(10). Soaps of monovalent cations form the typical oil-in-water emulsion, while soaps of divalent cations, such as calcium oleate, make the invert form of emulsion with oil as the external phase.

Parsons and Wilson(11) have shown the possibility of inversion of an emulsion by mixing solutions of sodium oleate in water with magnesium oleate in oil. The addition of di- and trivalent salts such as magnesium sulfate and ferric chloride inverted the oil-in-water emulsion, for instance. Our own experiments with calcium casein mixture[6] as the emulsifier have also shown the possibility of changing the type of emulsion by varying the proportions of oil to emulsifier.

Theoretically an emulsion with oil as the external phase would be more effective than one with water as the external phase, since then the active insecticide would come immediately into direct contact with the insect. Since, however, such emulsions cannot be diluted with water and are usually of such a tough, gummy nature that they cannot be broken up readily, they do not lend themselves to orchard practice.

Quick-Breaking Oil Emulsions.—The disadvantages of the oil-in-water emulsion have been overcome to a large extent by the

[6] The commercial mixture of powdered casein and hydrated lime used as an emulsifier is, in solution form, commonly spoken of as "calcium caseinate." This term will be used henceforth in this paper. The proportions of casein and lime are approximately 1 to 4.

development of a "quick-breaking" type of emulsion, which allows the water to separate out immediately on contact and run off, leaving a film of pure oil on the leaf surface. This brings the active insecticide, oil, instead of water or a hydrated colloidal solution, into direct contact with the insect. Neither water nor a hydrated colloidal solution has any practical insecticidal value. The "quick-breaking" emulsion thus increases the insecticidal action to such an extent that the almost prohibitive cost for an effective stable emulsion made from such highly refined lubricating oils as Oronite Crystal, is reduced to a point where these oils are economically practicable for orchard spraying.

The type of oil emulsion generally used in insecticidal work is that in which the oil is broken up into the smallest possible globules and distributed uniformly throughout the water. When this is accomplished, and the oil remains thus dispersed for an indefinite period of time without separation, the resulting mixture is known as a "stable" emulsion. If there is a tendency in the course of a short period of time for the oil to separate from the mixture, the emulsion is known as "unstable." Within certain limits this instability varies inversely as the percentage of the emulsifying agent.

In practice it appears that the strength of the interfacial membrane which separates the two phases of an emulsion varies a great deal, according to the emulsifier used. Some, such as are formed by "sodium-fatty-acid" soaps, are apparently very elastic and tough; others, such as are formed by typical colloids, as, for example, starch or colloidal copper, and also by calcium caseinate, are relatively very weak and easily disrupted.

In accordance with the general principle that the stronger the interfacial membrane the more stable the emulsion, it follows that the emulsifying agent which produces the weakest possible interfacial membrane is the best from the insecticidal standpoint. On this basis casein is better than soap and a metallic colloid is better than casein.

Some emulsifiers, such as lime or kaolin and other earths, are capable of absorbing considerable amounts of oil, as well as emulsifying them. This is particularly pronounced and important when the emulsifier or spreader is used in large quantities. All such absorbed oil is unavailable for liberation as a free liquid and constitutes, therefore, a permanent loss—assuming, of course, that free oil is the effective agent. Hence, from a theoretical standpoint, the use of the emulsifier which has the least possible oil-absorptive capacity is advisable (other things being equal). Colloidal copper is an almost ideal substance in all these respects, and much superior to typical

soaps, calcium caseinate, and lime. While we regard colloidal copper as a theoretically better emulsifier than calcium caseinate, the former in a pure state requires such care in its preparation and is so difficult to buy that it is impracticable for any but laboratory work. Calcium caseinate, however, is a widely distributed commercial preparation, and hence was chosen as the emulsifier for our experimental work.

In a stable emulsion of oil in water, the oil itself cannot come into contact with the object sprayed until separation of the two phases takes place, which in a highly stable emulsion may not occur except as the water disappears by evaporation. This obviously means a greater or lesser delay before the insecticidal (particularly wax-solvent) activities of the oil can begin. Secondly, it means a large loss of oil contained in the unavoidable "drip" or "run-off" from the sprayed surface. Thirdly, it appears reasonable to suppose that the interfacial film of emulsifier will be deposited as a more or less definite layer of inert substance between the oil and the leaf or fruit surface in such a way as to delay, even where it is not sufficient to prevent, the insecticidal action of the oil. This type of action would be especially important with typical "sodium-fatty-acid" soaps.

Lime, by its absorptive capacity, tends to prevent the oil from coming into direct contact with the object sprayed, and hence serves as an inhibiting factor. If we assume, therefore, that pure oil is the effective agent, it follows that the more "stable" the emulsion or the greater the absorptive capacity of the emulsifier the less value it possesses as an insecticide. This point was brought out early in 1925 by deOng and Knight(12) in a preliminary note based upon this project.

Most of the emulsions now on the market use various "sodium-fatty-acid" soaps as the emulsifying agent. While soap itself, owing to its fatty-acid content, is a weak insecticide, and may be fairly effective against soft-bodied insects like aphis and young scale insects, it is almost certain that when used as an emulsifier it is probably never in concentration sufficiently strong to be independently effective. This was demonstrated in our laboratory work when very strong solutions of many different soaps applied as sprays failed to affect a satisfactory kill of red scale, which is an armored species.

LABORATORY EXPERIMENTS WITH OIL EMULSIONS

For the purpose of routine insecticidal tests in the laboratory, the red scale, *Chrysomphalus aurantii* (Maskell), of the strain which has developed a resistance to HCN fumigation under orchard conditions, was chosen. So far as known, this is the most difficult of all citrus scales to kill by spraying, and it was assumed that if a spray could be developed which would kill this species, it would be effective against any of the others. A spray which apparently fulfills this requirement has been developed. From data thus far obtained it seems to be equally effective against the black and purple scales. This spray, however, has not yet had wide enough testing in the field to justify recommendation by this station for general use. Furthermore, as has been shown, certain peculiar effects are often produced upon the tree, which are not yet sufficiently well understood. For this spray a "neutral" white lubricating oil (Oronite Crystal oil, specific gravity 88, viscosity 106) was taken as the insecticidal agent, calcium caseinate (see p. 361) being selected as the emulsifier.

Lemons heavily infested with scale were used in the laboratory tests. Spraying was done by means of a small atomizer, and counts for determination of scale kill were made from ten days to two weeks after the application of the spray. The scale-infested lemons were hung in the laboratory during the interim.

The inhibiting effect, previously noted, of excess emulsifier was particularly well shown in an experiment wherein the amount of oil was maintained constant at 2 per cent, while the emulsifier was progressively reduced from 2 per cent to .0078 per cent (or from equal parts of oil and emulsifier to a ratio of 100 parts of oil to 0.39 parts of emulsifier). The killing efficiency was markedly accentuated as the amount of emulsifier was decreased. This is shown in table 3.

The natural mortality on checks kept under the same conditions was 49.7 per cent, or practically the same as lot 2 in table 3. The mortality was somewhat higher in lot 1 because one of the lemons had dried out. Desiccation has of itself a marked effect on the mortality of scale insects.

In the first three lots there were many live young present after treatment. Some were crawling about over the fruit while many others had just settled. Not until an oil film was formed over the surface of the fruit, was there any marked rise in the mortality. When this did occur the kill quickly rose to 100 per cent. It would seem, therefore, that the insecticidal agent is the free oil.

It is customary to use a spreader (generally calcium caseinate or glue) with many oil sprays for citrus trees. But a spreader is also an emulsifier, and tends further to inhibit the action of the oil through increased stabilization of the emulsion. Also, as in the case of certain emulsifiers such as lime or calcium caseinate, a spreader may accentuate the loss of oil through its additional oil-absorptive capacity.

TABLE 3

Relation of Scale Mortality to Concentration of Emulsifier in a Two-Per-Cent Emulsion of Oronite Crystal Oil

No.	Concentration of calcium caseinate *Per cent*	Scale surviving at end of test* *Per cent*	Remarks
1	2	32.3	Many young alive. Oil absorbed by surplus emulsifier.
2	1	49.5	Many young alive. Oil absorbed by surplus emulsifier.
3	.5	38.3	Many young alive. Oil absorbed by surplus emulsifier.
4	.25	7.5	Sprayed surface slightly greasy. No young alive.
5	.125†	0.0	Oil film just visible on sprayed surface.
6	.0625	0.0	Oil film distinct on sprayed surface.
7	.031	0.0	Well developed oil film present.
8	.015	0.0	Well developed oil film present.
9	.0078	0.0	Well developed oil film present.

* The usual natural mortality, about 40 to 50 per cent, is necessarily included in the counts.
† This corresponds to 1 pound of calcium caseinate to 100 gallons of spray.

Nothing in common use will spread better than oil. Hence in a quick-breaking emulsion a spreader is not needed. The spreading of any liquid is facilitated by reduction in its surface tension. To make oil spread better would therefore require the introduction of an oil-soluble constituent which would reduce the surface tension of the oil itself. Casein and all other such substances customarily used as spreaders are water-soluble, being practically insoluble in oil. Hence any "spreading effect" which results concerns the water alone and not the oil. This confusion has arisen from the mistake of considering an oil emulsion as a "solution" instead of as a mixture of two independent liquids.

The use of mechanical mixtures would evidently overcome this inhibiting effect of emulsifying agents. The method is not used because reliance cannot be placed upon the mechanical agitators at present in common use. But by very slightly emulsifying the oil, ordinary spray tank agitation is capable of overcoming the natural

buoyancy of the separated oil droplets and of maintaining a fairly uniform suspension of oil throughout the body of the liquid. The emulsifying agent is used in quantities just sufficient to separate the oil into relatively large droplets. The interfacial membrane is consequently weak and easily broken, thus liberating the enclosed oil. There is no danger that the stability of the system will be sufficient to withstand rupture upon impact with the leaf or fruit surface, and the maximum amount of oil is consequently freed and made available. On the other hand, the oil in the tank is maintained in the form of isolated droplets, there being no continuous sheet of oil to be broken up as would be the case with a mechanical mixture.

It has been found by observation that the individual globules of oil in the type of emulsion just described vary from about 0.1 mm. to 2.5 mm. in diameter, the smaller sizes largely predominating. The presence of oil droplets of the size indicated makes possible a very quick liberation of oil from the emulsion stage, while separation is very much slower in the stable type of emulsion made of extremely minute droplets. If the drops were uniformly 1 mm. in diameter, then in a 2-per-cent emulsion, 1 cc. would contain about 40 of these globules. Now, if 1 cc. be sprayed on a flat surface at a distance of 3 feet with an ordinary atomizer it will cover a circular area about 15 inches in diameter and the oil droplets will strike at widely separated spots, resulting in a typical "shotgun pattern." To form a film of oil over a given area, enough of the emulsion must be applied so that the droplets of oil will coalesce and form a film. In other words, the time spent while spraying becomes an important factor in application, for it is possible to increase the amount of oil on a surface by long or repeated sprayings. This factor varies with pressure, size of nozzle opening, and rate of discharge.

It has been found in practice that a 2-per-cent emulsion works very well in the field. If less than 2 per cent of oil is used, complete coverage will not be obtained without the use of excessive time in spraying. If more than 2 per cent is used there may be an undue accumulation of oil on the tree. In these sprays, raising the percentage of oil results merely in the liberation of more oil in the same period of time. The chief need in sprays of this type is the formation of a film of oil over the entire surface of the plant and the insect.

In the following test the emulsifier was varied from 5.0 to 0.0078 per cent while the oil remained constant. The emulsion was sprayed on a glass surface, the "run-off" collected and a quantitative determination of the oil present made.

Thus, as shown in table 4, the quantity of oil in the "run-off" from highly stable emulsions distinctly increased over that of the

original concentration, so that the recovered drip was actually richer in oil than the original spray. It is only with quick-breaking emulsions that the oil percentage in the drip falls markedly below the original concentration. In the best of these, which corresponds to the one adopted for our general work, the drip even then contains 0.34 per cent of oil or about one-sixth of the original amount.

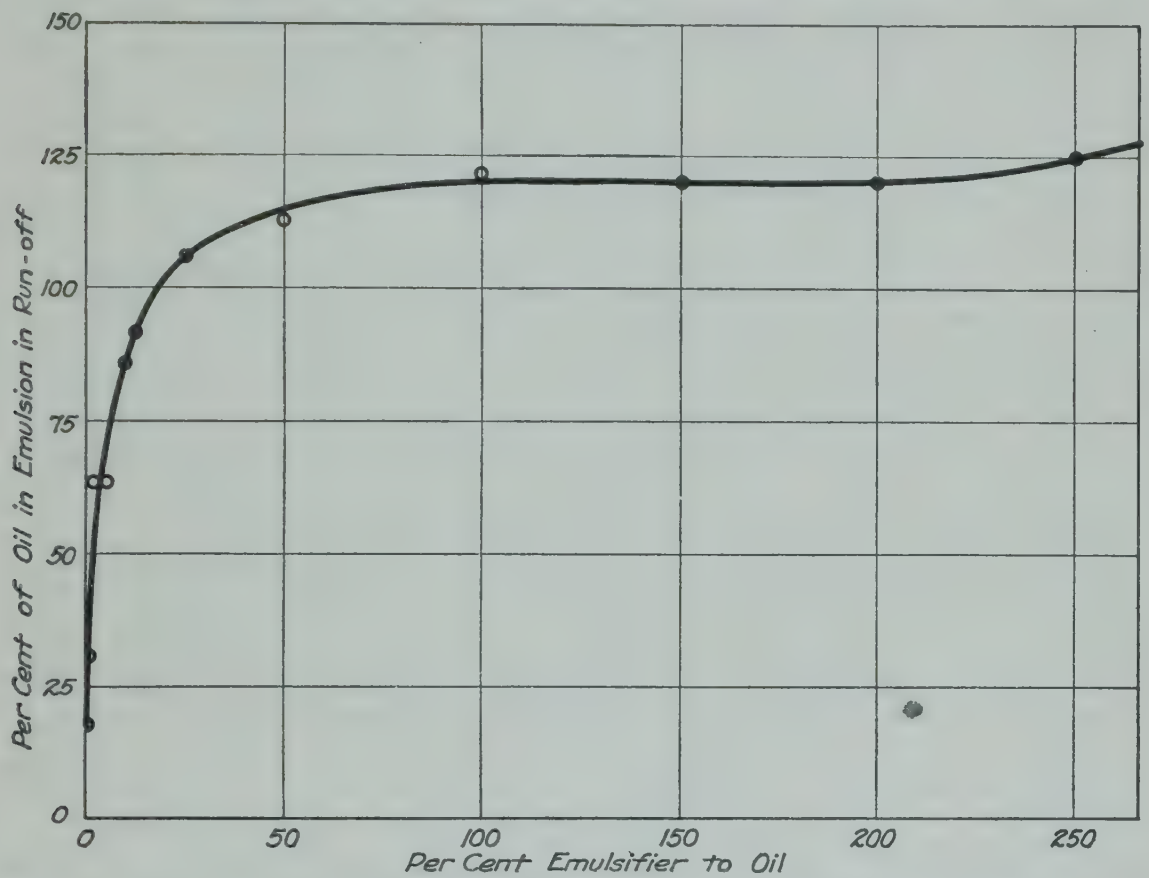

Fig. 2. Relation between amount of emulsifier used and the concentration of oil in the run-off from two-per-cent emulsions.

In experiments to obtain further evidence on the conditions of stability, emulsions were made as usual with Oronite Crystal oil. The standard emulsifier used was a mixture of powdered casein, selected for high solubility, and hydrated lime, the proportions being 1 to 4. Soap used in varying proportions as the emulsifier gave essentially similar results and hence these additional detailed data are not given.

A brief study brought out the following points in this connection. A stable emulsion containing 2 grams of oil to 98 grams of water was produced, (1) when 0.4 to 0.6 per cent of the 1 to 4 mixture of casein and hydrated lime was used; (2) when casein, dissolved in an amount of sodium hydroxide giving a hydroxyl concentration equal to that of the hydrated lime used in (1), amounted to 0.1 per cent, which corresponds approximately to 0.4 per cent of the 4 to 1 calcium casein mixture; and (3) when hydrated lime without casein was present in

TABLE 4

AMOUNT OF OIL IN RUN-OFF FROM A TWO-PER-CENT EMULSION MADE WITH VARYING AMOUNTS OF EMULSIFIER

Formula number	Calcium casein mixture *Per cent*	Ratio of emulsifier to oil *Per cent*	Ratio of concentration of oil in run-off to concentration in emulsion *Per cent*	Oil in run-off *Per cent*
1	5.0000	250.00	125.0	2.50
2	4.0000	200.00	120.0	2.45
3	3.0000	150.00	120.0	2.40
4	2.0000	100.00	122.0	2.44
5	1.0000	50.00	112.0	2.30
6	0.5000	25.00	106.0	2.20
7	0.2500	12.50	92.0	1.82
8	0.2000	10.00	86.0	1.70
9	0.1000	5.00	63.0	1.32
10	0.0500	2.50	63.0	1.24
11	0.0310	1.50	31.0	0.64
12	0.0078	0.39	17.0	0.34

the proportion of 0.6 to 0.8 per cent. The casein was found to be the more active emulsifying agent but the lime increased the general resulting stability. The principal value of the lime is in forming an alkaline solution, since casein dissolves in an acid or alkaline medium but not in a neutral one. A slight excess of lime also aids in neutralizing some of the soluble salts found in water which might hinder emulsification. Ordinary soap is now seldom used in spray practice for making emulsions, because the sodium base reacts with the calcium and magnesium salts in solution in many waters, forming insoluble soaps such as calcium oleate, thus causing the emulsion to break prematurely.

It was found, as the result of laboratory spray tests, that Oronite Crystal oil could be used effectively in emulsions of the quick-breaking type at two-per-cent concentration.

The following formula was finally developed as a standard for both laboratory and field use. The percentage values are obviously only approximate:

Oronite Crystal oil 2 per cent (2 gallons)
Calcium caseinate 0.0078 per cent (28.3 grams or 1 ounce, approximately)*
Water 98 per cent (98 gallons)

* As a result of field experience during the two years since this manuscript was originally prepared for publication it has been found that owing to the very general inefficiency of the average spray tank agitator, it is best and safest to use two to three times this quantity of emulsifier. This is not sufficient to noticeably affect insecticidal results, at least in the field.

In this formula the calcium caseinate is present in the proportion of 1 part to 200 parts of oil. In the laboratory the calcium caseinate is first dissolved in the water, then the oil is added and the whole violently shaken in order to produce emulsification. In the field a slightly different procedure is necessary. The calcium caseinate is first completely dissolved in about a quart of water. It is then added to from twenty-five to fifty gallons of water in the spray tank. The oil is next added and the agitator is started at the same time. The tank is then filled with water while the agitator is running. The emulsion is then ready to apply.

There is one point peculiar to this type of spray which is of considerable practical importance. The oil droplets are large and highly buoyant, and therefore quickly float to the surface of the water and form a definite layer or sheet of oil, which is, however, still emulsified. Vigorous agitation is, therefore, required to keep the spray of uniform consistency throughout. Only spray rigs which possess the most efficient type of agitator should be used to apply oil emulsions as quick-breaking as the one described above.

This spray has given 100 per cent kill of resistant red scale *in the laboratory*, where every scale insect was actually treated. In the field, owing to the impossibility of complete coverage and the possible effects of other factors this efficiency has never been attained.

FIELD TESTS OF A QUICK-BREAKING EMULSION

The quick-breaking Oronite Crystal oil emulsion previously described has been tested in the field. The formula found most satistory in the laboratory tests was used. As previously indicated, the resulting kills have never been as efficient(as might be expected) as those attained in the laboratory work. The results are shown in table 5.

These kills resulted from very careful application. The percentage surviving from average commercial spraying with the same material would no doubt often be above these figures. The results given are based on counts made on the fruit. Less satisfactory results occur on the twigs, possibly because of their greater oil-absorptive capacity, thus resulting in a less permanent oil film.

The criterion of effective application is complete coverage resulting in the presence of a visible film of oil over the entire surface of the plant, after the water carrier has evaporated.

TABLE 5

MORTALITY OF RED SCALE IN FIELD TESTS OF A QUICK-BREAKING TWO-PER-CENT EMULSION OF ORONITE CRYSTAL OIL

Place	Per cent of scale† surviving on fruit at end of test
Riverside, Calif.	5.05 and 2.94 (2 plots)
Whittier, Calif.*	7.95
La Habra, Calif.*	2.96
Santa Ana, Calif.*	2.13 (On purple scale, *Lepidosaphes beckii* (Newman), 2.00)
Tustin, Calif.*	3.43
Santa Barbara, Calif.	5.86 and 3.90 (2 plots)
Lindsay, Calif.	7.8 and 0.0 (2 plots) (on the citricola scale *Coccus pseudomagnoliarum* Kuwana)
Average	4.0

* Resistant-scale areas.

† The red scale *Chrysomphalus aurantii* (Mask.) is meant except as otherwise noted.

EXPERIMENTS RELATING TO THE NATURE OF THE INSECTICIDAL ACTION OF NEUTRAL OILS

The following test illustrates the essential blandness and "neutrality" characteristic of these white, highly refined petroleum oils, a fact which finds further confirmation in that it is this type of oil which is utilized in human medicine. Coleman's mealybug (*Phenacoccus colemani* Ehr.) were continuously immersed in Oronite Crystal oil and examined twice a day until all had died. Death was assumed to take place concurrently with cessation of all bodily movements, as determined by absence of response to stimulation by a needle.

Table 6 includes the combined results of two distinct tests. The same data are shown graphically in figure 3.

Some supplementary data on other insects were obtained which check very well with the results recorded above.

Thus with cabbage aphis (*Aphis brassicae* Linn.), out of eight individuals which were tested two were still alive after 18 hours' immersion in the oil. There is little doubt that aphids on the whole are more susceptible than mealybugs.

Larvae of the orange tortrix (*Tortrix citrana* Fern.) likewise survive a considerable period of immersion in this oil. In one test involving two individuals, one specimen was dead at the end of the 72nd hour and the other at the end of the 96th. In this case cessation of the pulsation of the dorsal vessel was taken as indicative of death.

In the case of ladybird beetles (*Hippodamia convergens Guerin*) visible movements cease in from 3 to 7 minutes.

TABLE 6

SURVIVAL OF COLEMAN'S MEALYBUG IMMERSED IN ORONITE CRYSTAL OIL

Hours elapsed since beginning of test	Insects dead	Insects still living
0	0	40
68	1	39
72	6	33
77	4	29
96	1	28
114	4	25
120	5	20
144	2	18
148	1	17
160	1	16
166	1	15
184	4	11
240	3	8
264	1	7
288	2	5
294	2	3
312	1	2
336	1	1
384	1	0
Totals (end of test)	40	0

Average time of lethal immersion, $\frac{6524}{40} = 163$ hours, or nearly 7 days.

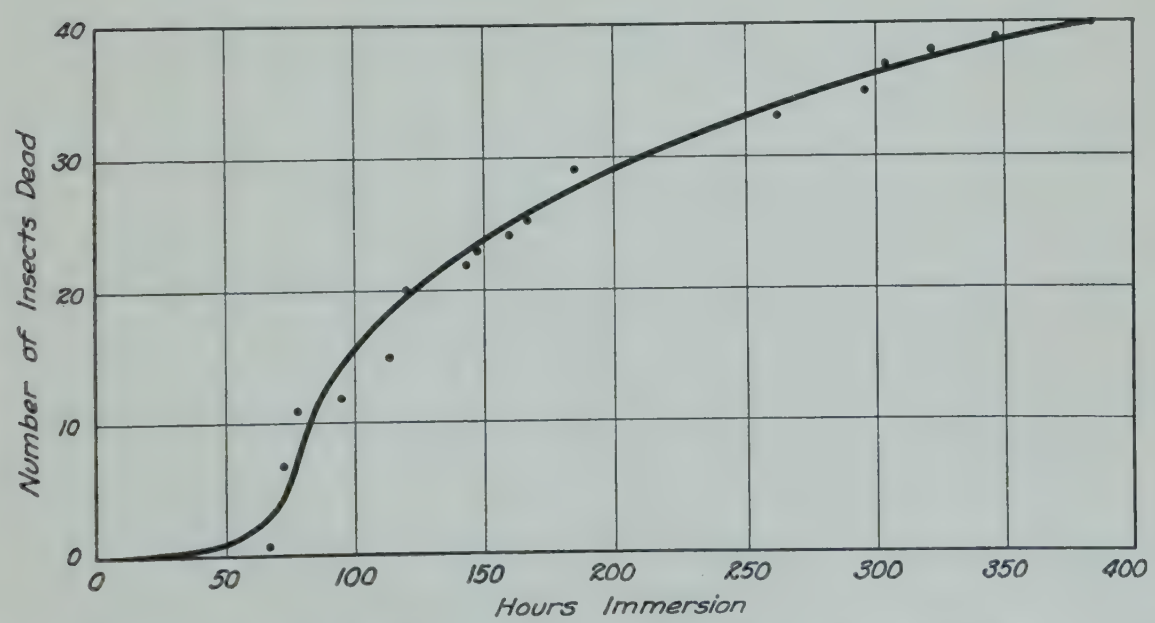

Fig. 3. Lethal curve of Coleman's mealybug, *Phenacoccus colemani* Ehrhorn, immersed in Oronite Crystal Oil.

The contrast between the long period of immersion required for mealybug and the shorter one for beetles was puzzling. As a check, beetles were immersed in tap water, and the apparent anomaly was then explained. Furthermore, there were obtained some data confirming our belief that the lethal effects of these highly refined oils could be explained almost solely upon the basis of suffocation.

Beetles were floated to the top of a water-filled, inverted test tube, and it was found that all visible movements ceased within practically the same length of time as in the oil. That cessation of movement in this instance was not indicative of death was clearly shown when beetles that had been immersed in water for several hours revived very rapidly upon being warmed and dried. This indicates that with these insects at least, cessation of movement is not a definite criterion of death.

There is little doubt that oil-immersed beetles would likewise revive rapidly and completely if the adhering oil could be dissipated as completely and rapidly as the water. The fact that they do not do so indicates that enough oil adheres permanently to the body and completely covers the spiracles so that the insect cannot be removed from its oil bath.

Mealybugs were likewise treated with water as a lethal agent. Four insects so immersed for four hours and apparently dead, all movement having ceased, revived completely. In a succeeding check test ten mealybugs were kept under water for a period of five hours. Of these only two revived. These results indicate a very much lower average lethal immersion limit for water than for oil. Movement ceases much sooner in water than in oil.

As a final test of the "oxygen-deprivation hypothesis" eighteen mealybugs were placed in an atmosphere of pure hydrogen, which is essentially inactive so far as living organisms are concerned. Impurities due to the processes of generation were doubtless present in some degree, but on the whole the results check very well with those previously given. Of the eighteen specimens treated eleven were dead at the end of 24 hours; four more at the end of 48 hours and the remainder (thirteen) at the end of 72 hours. This is an average lethal immersion period of 64 hours, approximately two and one-half days.

In view of the foregoing data, it may be stated that death of scale insects through the action of white neutral oils may be ascribed almost entirely to suffocation. At least, this one factor offers a satisfactory explanation for all the known facts.

After the completion of the original draft of the manuscript of this paper it was found that we had overlooked two important articles bearing upon this same subject, written by George D. Schafer (13, 14) in 1911 and 1915 respectively. It is unnecessary to review his conclusions relative to our work, which was done entirely independently, but it is worth while to note that our results bear practically the same implications and are confirmatory of his conclusions on the subject of oxygen deprivation.

TABLE 7

Relation of Viscosity of Oil to its Effect on Red Scale

No. of test	Oil description	Viscosity	Percentage of oil in emulsion	Per cent scale surviving test*	Remarks
1	Castor oil	1840	2	48.0	No more than natural mortality.
2	No. 6, a heavy lubricating oil	364	2	0.0	
3	Oronite Crystal oil	100	2	0.0	
4	A special light lubricating oil (specifications not given)	38	2	2.0	This oil was just below the lethal viscosity limit.
5	No. 4a, a refined kerosene	21+	20†	19.0	

* The basis for scale counts ranged from 200 to 600 insects.

† In spite of the high percentage here used only a very poor kill was obtained; at 2 per cent only the natural mortality would have been found.

In laboratory tests, oils within a rather wide range of high viscosity, other things being equal, gave complete control. Below the minimum of this range, the lighter an oil the less certain will be the kill. On the other hand, extremely high viscosities are likewise ineffective. These facts are illustrated in table 7, which is based upon laboratory tests.

Under the heading "viscosity" are given a series of values which are approximate only. Oronite Crystal oil was arbitrarily taken as a standard and assigned a value of 100. The values were determined by measuring the time of flow of 50 cc. of the oil from a small burette at a constant temperature.

These tests were made with the resistant red scale (*Chrysomphalus aurantii*). All emulsions were of the quick-breaking type.

These oils (excepting castor oil) are all almost entirely non-toxic, and castor oil even with its toxicity fails to kill. Evidently castor oil and oil 4a are ineffective for different reasons. In the case of castor oil the cause is probably mechanical, as this oil is apparently too viscous to spread evenly and form a continuous film. In the case of oil 4a, on the other hand, the oil evaporates so quickly that a film is not maintained long enough to kill the more resistant individuals. The minimum viscosity[7] limit for complete killing evidently lies somewhere between Oronite Crystal oil and the "special light lubricating oil" used in this test.

Dilution tests (table 8) were then made and found to conform in general to the conclusion just stated. Kerosene distillate was used in making these viscosity reductions. These tests are not conclusive and must later be greatly extended, particularly toward the lower limits.

TABLE 8

Effects of Diluting Heavy Oils with Kerosene Distillate

Oil	Viscosity	Percentage of oil in emulsion	Percentage scale surviving test
Castor oil plus kerosene distillate	74±	2	0.0
Oronite Crystal oil plus kerosene distillate	45±	2	0.0
Oil 6 plus kerosene distillate	100	2	0.0

Kerosene distillate is of itself ineffective, but when its viscosity is increased by the addition of castor oil (or vice versa) a kill is immediately obtained. Toxicity is of paramount importance in assigning practical limits to degrees of volatility (viscosity) permissible in a given oil. For instance, an oil which might be volatile enough to disappear completely in one hour, if also sufficiently toxic to penetrate and kill the most resistant individual scales treated in thirty minutes, would obviously be entirely effective as a spray material. On the other hand a non-toxic oil which would volatilize completely in ten days would not suffice to kill red scale. The importance of these two factors lies not so much in their *absolute* as in their *relative* values.

[7] The terms 'low viscosity' and 'high volatility' cannot be used interchangeably in all cases. Oils from a similar source, distilled at the same range of temperature will be quite uniform in viscosity, but in the process of refining, the viscosity changes enormously, while volatility may remain constant. The blending of oils of different viscosities may also destroy the correlation between viscosity and volatility.

PRELIMINARY TESTS OF TOXICITY OF INSECTICIDAL MATERIALS

Table 9 gives data relating to the toxicity of a series of oils and other substances. These values should be self-explanatory in view of the preceding discussion. The tests were not all made in the same way and on the whole can be relied upon to give an idea of relative toxicity, but not of the minimum lethal limit, which is ultimately the most important factor. Five mealybugs, *Phenacoccus Colemani* (Ehrhorn), were used in making each of the determinations. Substances are listed according to toxicity, the more toxic ones coming first.

The possibility of imparting toxicity to otherwise neutral oils through the addition of toxic constituents (fatty acids or unsaturated hydrocarbons, for instance) and hence permitting higher volatility and shortening the time of insect kill may be of great importance in future work. This raising of the volatility is also of considerable significance in decreasing plant injury. Long persisting oils may tend to upset the metabolic processes of the plant even where no immediate effect is noticeable.

TRACHEAL PENETRATION OF INSECTICIDES AND SIGNIFICANCE OF SOLUBILITY OF WAX IN OILS

A study was made of the penetration of different fractions of petroleum oils, some of the vegetable oils and other spray materials, into the tracheal system of the red scale. This work was somewhat similar to that of Moore (7) on tracheal penetration.

For this purpose specimens were chosen that had passed through the second moult but had not yet reached maturity. At this stage of development the insect is free from the scale covering and can be lifted out intact. The detached insect is placed on a slide, ventral side up, and when it is immersed in liquid the tracheal system becomes plainly visible. The low refractive index of the air-filled trachea causes them to show as black lines under the microscope. If penetration of the liquid occurs, it causes an increase in the refractive index of the liquid-filled portion with a consequent lowered visibility, and the degree of penetration becomes plainly visible.

Figure 4 is a photomicrograph showing the main branches of the tracheal system of the red scale. It will be noted that the spiracles

TABLE 9

RELATIVE TOXICITY OF INSECTICIDAL SUBSTANCES TO MEALYBUGS AS INDICATED BY PERIOD OF LETHAL IMMERSION

Rank	Substance	Time of lethal immersion	Remarks
1	Benzol	3 seconds	Chemically pure
2	Ether	10 seconds	Chemically pure
3	Grain alcohol	90 seconds	95 per cent pure
4	"Zero" rosin oil	3 minutes	Georgia Rosin Products Co.
5	Double Run Zero rosin oil.	3 minutes	Georgia Rosin Products Co.
6	Triple Zero rosin oil	3 minutes	Georgia Rosin Products Co.
7	Turpentine	3.5 minutes	Commercial
8	Double-distilled coconut fatty acid.	6 minutes (average)	Armour & Co.
9	London rosin oil	7 minutes	Georgia Rosin Products Co.
10	Oleic acid	7–12 minutes	Commercial
11	Shale oil	12 minutes (maximum)	California distilled
12	Petroleum oil 1†	17 minutes (maximum)	Standard Oil Co. of Calif. (lubricating-oil distillate)
13	Furfural	10–13 minutes	Insects only partially immersed, vapors evidently toxic.
14	Special "X" rosin oil	20 minutes (average)	Georgia Rosin Products Co.
15	Liquid asphalt	30 minutes*	Standard Oil Co. of Calif.
16	Unsaturated hydrocarbons removed from kerosene	43 minutes (average)	Standard Oil Co. of Calif.
17	Vaseline plus water white distillate.	60 minutes*	Vaseline 1 part, kerosene 5 parts.
18	Petroleum oil 3†	60–360 minutes	Standard Oil Co. of Calif. (lubricating-oil distillate)
19	Petroleum oil 1a†	60–1200 minutes*	Standard Oil Co. of Calif. (kerosene distillate)
20	Petroleum oil 3a†	108 minutes (average)	Standard Oil Co. of Calif. (kerosene)
21	Petroleum oil 2a†	120 minutes*	Standard Oil Co. of Calif. (kerosene)
22	Petroleum oil 4a†	120 minutes*	Standard Oil Co. of Calif. (the least toxic of the kerosenes)
23	Whale oil	210 minutes*	Crude. Highly toxic.
24	Petroleum oil 2†	240–720 minutes	Standard Oil Co. of Calif. (lubricating-oil distillate)
25	Cottonseed oil	13–1400 minutes	Crude

* The definite meaning (whether average, maximum or approximate survival limit) of the value given is unknown.

† See table 1 for further specifications.

TABLE 9—(*Continued*)

Rank	Substance	Time of lethal immersion	Remarks
26	Linseed oil	30–1300 minutes	Commercial
27	Petroleum oil 4†	240–1400 minutes	Standard Oil Co. of Calif. (lubricating oil)
28	Castor oil	1400 minutes*	Refined
29	Fish oil	1400 minutes*	Commercial
30	Petroleum oil 4x†	1400 minutes*	Standard Oil Co. of Calif. (lubricating oil)
31	Olive oil	2500 minutes*	Refined
32	Petroleum oil 6†	2500 minutes*	Standard Oil Co. of Calif. (lubricating oil)
33	Petroleum oil 5x†	2500 minutes*	Standard Oil Co. of Calif. (lubricating oil)
34	Petroleum oil 7†	1200–5760 minutes	Standard Oil Co. of Calif. (lubricating oil)
35	Petroleum oil 5† (Oronite Crystal oil)	9780 minutes (average)	Standard Oil Co. of Calif. (lubricating oil)

* The definite meaning (whether average, maximum or approximate survival limit) of the value given is unknown.

† See table 1 for further specifications.

are connected with each other by four large tracheal trunks. In addition a large number of smaller branching tubes ramify to all parts of the body. For purposes of comparison the spiracle-connecting trunks of the tracheal system are divided into three areas designated as A, B and C.

That portion of the trunk extending from the opening of the spiracle to the point where the trachea first branches is designated A. About one-third of the distance between the spiracles along the main tracheal trunk is designated B, and the entire distance C. In figure 4, these distances are shown from one spiracle only. When the oil penetrates far enough to fill the four trunks it has usually, at the same time, completely filled the smaller branches.

Table 10 shows that neither lime-sulfur solution (in ordinary dilution) nor Bordeaux 5–5–50 showed any penetration at all. When lime-sulfur and a proprietary miscible oil were combined there was a fair degree of penetration. All emulsions of the lubricating-oil type, as well as the lubricating oils themselves, gave good penetration, filling the entire tracheal system. The kerosenes, on the other hand, were very erratic in behavior. Initial penetration (2–8 minutes) was in nearly all cases very rapid but in a considerable number of instances this gradually ceased, and movement of the liquid was reversed so

that the tube emptied itself. Apparently the insects have the power of expelling these light oils from the tracheae. Furthermore, they are then able to keep these oils from penetrating for more than 30 to 40 minutes. This ability of the insect, particularly when taken

Fig. 4. Ventral aspect of the red scale, *Chrysomphalus aurantii* (Maskell), showing the tracheal system.

in connection with the high volatility of the oils, excludes the kerosenes from the class of satisfactory scalecides even though their toxicity is relatively high compared with that of neutral lubricating oils.

In these tests it was found that soaps, oils, stable emulsions and water-glue solutions were capable of penetrating into the tracheae

of these detached insects. In some cases, as in highly stable oil emulsions and lime-sulfur oil mixture, the penetration seemed to be that of the emulsion itself. This seems to raise a question, considering the emphasis previously laid upon the necessity of oil liberation. It is obvious that anything, whether a pure oil or an emulsion, which would completely clog the spiracles would ultimately suffocate the insect.

TABLE 10

TRACHEAL PENETRATION OF RED SCALE BY INSECTICIDAL SUBSTANCES

Sprays: Material	Tracheal penetration
Lime sulfur, 2 per cent	None
Lime sulfur, 10 per cent	None
Lime sulfur, 20 per cent	A
Lime sulfur, undiluted	A
Bordeaux, 5–5–50	None
Miscible oil, 2 per cent and lime sulfur, 1 per cent	B
Commercial oil emulsion, 5 per cent (standard type)	C
Fish-oil soap and Oronite Crystal oil emulsion, 6 per cent	C
Fish-oil soap and No. 6 petroleum oil emulsion, 6 per cent	C
Fish-oil soap and kerosene distillate (Oil No. 1a) emulsion, 6 per cent	Erratic, initially rapid
Fish-oil soap and kerosene oil No. 2a emulsion, 6 per cent	Erratic, initially rapid
Bordeaux and Oronite Crystal oil emulsion, 6 per cent	C
Ferrous sulfate and Oronite Crystal oil emulsion, 6 per cent	C
Oils (pure):	
Oronite Crystal oil (No. 5)	C
Petroleum oil No. 6	C
Kerosene, No. 2a	C
Kerosene distillate, No. 1a	C
Cottonseed oil (Crude)	C
Oleic acid	B
Turpentine	B
Linseed oil	C
Miscellaneous:	
Tap water	None
Sodium silicate (water glass)	A
Xylol stained with Sudan III	Erratic, ultimately complete
Xylol (pure)	C

The paradox is explicable, however, when we consider the penetration of the trachea of an insect *in situ*. Here the armored scale (red scale), which was the subject of discussion in connection with the liberation of oil from quick-breaking emulsions, is completely protected by both a dorsal and a ventral waxy scale covering. For the insecticide to come into actual contact with the spiracles necessitates first of all penetration of this scale covering. Water is not a wax solvent and is hence completely excluded from penetrating this covering. The same consideration applies to any water mixture, including stable emulsions wherein water is the continuous phase and where little or no oil liberation takes place. Free oils on the other hand are not only capable of tracheal penetration but are wax solvents as well and hence capable of penetrating the scale covering. In a stable emulsion the oil is kept largely locked up and hence can exert no independent effect upon the wax.

This explanation is borne out by the fact that miscible oils (as is found in current practice in the field) may be fairly effective against the unarmored black scale (*Saissetia oleae* Bernard), where the spray is able to gain unobstructed access to the tracheal opening, while they fail in large degree in the case of red and other armored scales.

VEGETABLE OILS

In addition to the petroleum oils, certain vegetable oils were tested as spray materials. These include cottonseed, linseed, castor, and rosin oils. These are all much more toxic to insects (as indicated in table 9) than neutral white oils, no doubt on account of their fatty-acid content. These fatty acids seem in general to correspond to the unsaturated hydrocarbons of the petroleum oils. Like the latter, they are toxic to plants as well as to insects, although in many instances at least, to a less degree.

The most promising of the vegetable oils tested was cottonseed oil. In the field this gave an excellent kill of scale, but defoliation was considerably more severe than was the case with Oronite Crystal oil. There is a great field for future investigation of vegetable oils in connection with insecticidal spray work.

COCONUT FATTY ACID

In view of the widespread interest in the recent development of the use of coconut-oil fatty acids by Siegler and Popenoe(15), it is well to call attention to the fact that while they undoubtedly have marked insecticidal properties, they are also exceedingly toxic to plant tissue when used in concentrations even remotely approximating those necessary to kill armored scale insects of citrus. The following test shows this conclusively.

An emulsion was made with equal parts of fatty acid and gasoline, as recommended in the reference quoted, but the amount of glue was reduced so as to make an emulsion of the quick-breaking type. This mixture was diluted with water until the emulsion contained 2 per cent of the fatty acid. A potted citrus plant was sprayed with this emulsion with disastrous effect. It was completely defoliated and both the leaves and the twigs were badly burned and spotted.

Lemons infested with red scale were sprayed with the same emulsion. The fruit was burned and shriveled, but only 97 per cent of the scale was killed. Furthermore, at the time the examination was made (ten days after spraying) young were hatching and settling on the fruit. In this instance, although severe injury resulted to the plant and fruit, the scales were not all killed. This failure is probably due in large part to lack of wax solubility, with consequent inability to penetrate the scale covering. This material would probably be much more effective against the unarmored black scale (*Saissetia oleae*).

On diluting the emulsion one-half, so that it contained 1 per cent of fatty acid, a mortality of only 83.4 per cent was found and the fruit was again shriveled.

The test was repeated without the gasoline in order to get the full effect of the undiluted fatty acid. An application of a two-per-cent emulsion left only 0.66 per cent of the scale alive but severely injured both the fruit and plant.

Finally a stock emulsion was applied, made according to the formula recommended, with its full complement of glue (giving a stable emulsion) and containing 2 per cent of fatty acid (or 80 times as strong as recommended for aphids). No injury to the plant resulted from this application. When the determination of insect mortality was made, there could be found no indication that an insecticide had been applied, the count showing mortality of 43.6

per cent, which is equivalent to natural mortality only. The spray had proved totally ineffective. Young scale were found crawling freely about within two days after the application.

These tests again confirm the conclusions previously reached regarding the necessity of a quick-breaking emulsion in order to liberate the insecticidal agent for effective use.

SUMMARY

1. Petroleum oils of the kerosene and "stove-distillate" type (28°–32° A.P.I.) have been occasionally used as insecticides over a period of many years in citrus orchards, with varying results as to insecticidal effects and injury to the tree and fruits.

2. Non-viscous oils of a low boiling point, such as the kerosenes, are safer to use on the tree than those of high boiling points, but are unsatisfactory as scalecides because of relatively low toxicity combined with high volatility.

3. Highly refined, white lubricating oils are probably the most advisable for use on citrus trees, especially at summer temperatures. Oils of a low viscosity are apparently safer to use on trees than those of high viscosity. This is due to the more rapid disappearance of the former.

4. Severe injury to the citrus trees from the use of lubricating oil is associated with the presence of a high percentage of unsaturated hydrocarbons. Refining petroleum oil with sulfuric acid removes the following injurious constituents: aromatics, olefins, resins, and sulfur.

5. The filtration of petroleum oils through Fuller's earth has not shown itself effective in reducing the amount of injurious constituents present.

6. Gross symptoms of injury to citrus trees from the use of unrefined petroleum oils, include defoliation, fruit spotting and dropping and the killing of twigs and branches. In addition to these injuries, there is an apparent interference with the normal plant functions of transpiration and respiration.

7. A quick-breaking emulsion utilizes to the maximum degree the insecticidal agent. Two per cent non-volatile lubricating oil with 98 per cent of water as a carrier has, when applied as a quick-breaking emulsion in the laboratory, produced a complete kill of red scale on lemons. Stable oil emulsions using the same ingredients are ineffective against this scale at strengths of from 4 to 8 per cent actual oil.

8. The "quick-breaking" action in an emulsion is greatest when the average size of the dispersed oil globules is greatest, and that size is greatest when the proportion of emulsifier to oil is least.

9. The concentration of oil in the run-off from sprays containing 2-per-cent concentration of oil varied from 2.5 per cent for the stable type of emulsion to 0.39 per cent for the quick-breaking type in laboratory tests on glass plates.

10. The insecticidal action of unrefined lubricating oils seems to be the result of two principal lethal factors. These are suffocation and toxic action, or poisoning. The former results chiefly from non-volatility (film permanence), the latter chiefly from the action of unsaturated hydrocarbons in the case of unrefined petroleum oils, or that of free fatty acids in the case of vegetable oils.

11. The wax solubility of oils is one of the important factors determining the insecticidal effectiveness of lubricating oils against the red scale. In the quick-breaking emulsions the free oil dissolves the waxy scale covering and enables the oil to penetrate to the spiracles; stable emulsions, with which the liberation of free oil does not readily occur, lack this feature to a great extent and are therefore not so effective.

12. The lethal immersion period varies from a few seconds for the most toxic substance tested to sixteen days for the least toxic.

13. Volatility limits of oil range from a few minutes or hours to several weeks.

14. Various physiological disturbances, which are highly characteristic and little understood, are induced in citrus trees by the use of "neutral" white oils in quick-breaking emulsions. These disturbances are evidenced by special types of leaf and fruit drop but not by actual burning or spotting (except possibly in rare instances).

15. Free fatty acids—while highly effective as insecticides for aphids—are not suitable for use in quick-breaking emulsions at the high concentration required for the control of scale insects, because of the injurious effects on plant tissue of such concentrations of the acids.

16. The present paper is published as a progress report on a special investigation, the results of which, while highly suggestive and important, should not be construed as constituting a recommendation for practical orchardists.

LITERATURE CITED

1. Quayle, H. J.
1922. Resistance of certain scale insects in certain localities to hydrocyanic acid fumigation. Jour. Econ. Ent. **15**: 400.

2. Gray, Geo. P., and E. R. deOng
1925. Laboratory and field tests of California petroleum insecticides. Indus. Eng. Chem. **18**: 175–180.

3. Volck, W. H.
1903. Spraying with distillates. California Agr. Exp. Sta. Bul. **153**: 1–31.

4. Magness, J. R., and A. M. Burroughs
1921–22. Apple storage investigations. Marble Lab., Canton, Pa., Rept.

5. Burroughs, A. M.
1923. Effects of oil sprays on fruit trees. Proc. Amer. Soc. Hort. Sci. **1923**: 269–277.

6. Edeleanu, L.
1914. Refining petroleum by liquidified sulfur dioxide. Amer. Inst. Min. Eng. Bul. **93**: 2313.

7. Moore, Wm.
1918. A study of the toxicity of kerosenes. Jour. Econ. Ent. **11**: 70–75.

8. Fawcett, Howard S.
1916. A spotting of citrus fruits due to the action of oil liberated from the rind. California Agr. Exp. Sta. Bul. **266**: 261–269.

9. Tinkle, P., H. D. Draper, and J. H. Hildebrand
1923. The theory of emulsification. Jour. Amer. Chem. Soc. **45**: 2780–2788.

10. Bhatnagar, S. S.
1921. Studies in emulsions. Jour. Chem. Soc. **119**: 61.

11. Parsons, Leon W., and O. G. Wilson, Jr.
1921. Some factors affecting the stability and inversion of oil-water emulsions. Jour. Indus. Eng. Chem. **13**: 1116–1123.

12. deOng, E. R., and Hugh Knight
1925. Emulsifying agents as an inhibiting factor in oil sprays. Jour. Econ. Ent. **18**: 424.

13. Schafer, Geo. D.
1911. How insecticides kill. Parts 1 and 2. Michigan Agr. Exp. Sta. Tech. Bul. **11**: 1–65.

14. Schafer, Geo. D.
1915. How insecticides kill. Part 3. Michigan Agr. Exp. Sta. Tech. Bul. **21**: 1–67.

15. Siegler, E. H., and C. H. Popenoe
1925. The fatty acids as contact insecticides. Jour. Econ. Ent. **18**: 292–299.

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EFFECT OF MULCHES ON SOIL TEMPERATURES DURING THE WARMEST WEEK IN JULY, 1925

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Previous investigations[1, 2, 3] on the use of paper as a mulch in contrast to soil mulches have dealt mainly with the effect on crop growth and crop yields. Investigations are now under way regarding the effects of such paper covering on the temperature and moisture conditions of the soil, and also on the most desirable kind of paper to use with particular regard to color, weight, durability, and need for perforations.

By the use of an unperforated black paper mulch at Berkeley in 1924, Shaw[4] found that the soil at a depth of three inches averaged about 0.42° F warmer than at a similar depth in the soil-mulched plots. Hartung[2] in the pineapple fields of Hawaii obtained higher mean soil temperatures at a depth of three inches in areas covered with grayish brown paper mulches than in unprotected soil. In his summary he states that "paper mulch maintains a mean soil temperature in the upper 3-inch layer of soil during the cool season in the localities given, from 3 to 4.5 degrees Fahrenheit above that of non-paper covered soil; provided the mulch paper is dark, preferably black in color." Although the standard mulch paper which Hartung used presented a "greyish brown appearance," he nevertheless recommends black paper. Stewart[5] and his co-workers in Hawaii found that on clear days the areas covered with black paper were from 12° to 15° F warmer during the day, and from 4° to 5° F warmer during the night.

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Fig. 1.—Soil temperature plots at Davis, California, January, 1925.

METHODS

The experimental tract used in these experiments is an area of Yolo loam which had been summer fallowed in 1924. The surface soil to a depth of three feet is a fairly uniform loam on a subsoil of fine sandy loam with some minor variations of coarser texture. This soil is of recent alluvial deposition, is derived mainly from sedimentary rocks, and occupies a nearly flat topographic position. Sixteen plots, each five meters square, were arranged in this area, separated by paths two meters wide.

A sixteen-point recorder was installed in a one-story frame structure, 150 feet north of the experimental area, in January, 1925, with all connecting wires carried to the plots in overhead conduits as shown

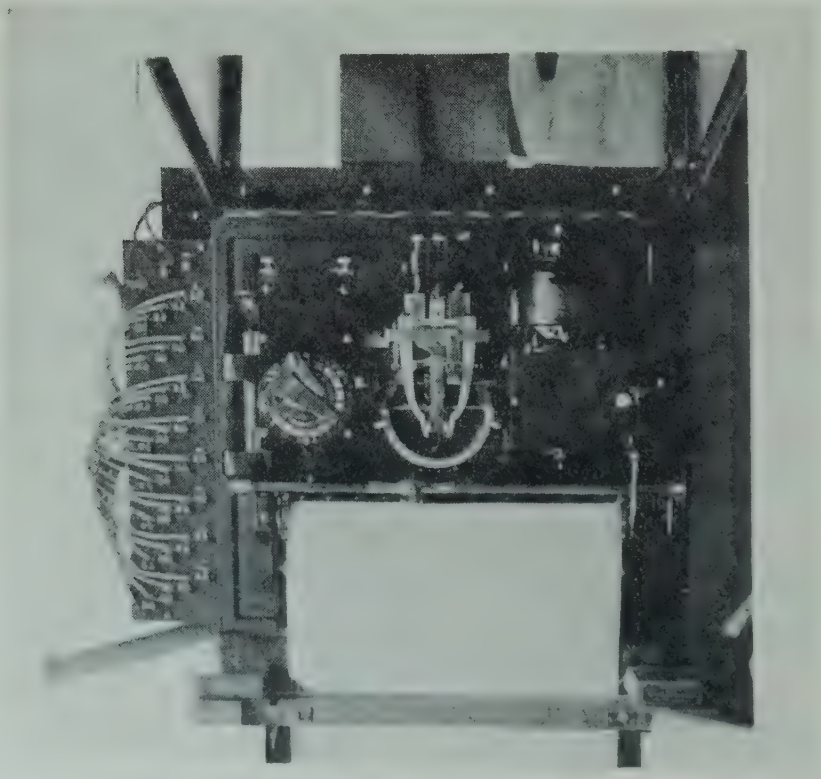

Fig. 2.—Automatic temperature recorder.

in figure 1. Sixteen resistance thermometers were carefully standardized against standard mercury thermometers and were placed in the soil at the center of only five of the plots at depths of from ½ inch to 36 inches. The least number of thermometers in any one plot was two and the most was six. In burying the thermometers, a hole of small diameter was dug, the soil of the various horizons being carefully laid aside. The thermometers were then inserted in the undisturbed soil on the north side of the holes, so that the resistance units or "bulbs" were at least eight inches from the excavation. The hole was then filled, the soil layers being put back in proper order and lightly tamped in order to attain the same degree of compactness as existed originally.

The temperature recorder (fig. 2), a Leeds & Northrup 16-point recorder, was adjusted so that the temperature of each individual thermometer was recorded every 15 minutes, giving 96 records in a 24-hour period or 10,752 records from the 16 thermometers in a period of 7 days. All temperatures herein reported are in degrees Fahrenheit.

A continuous record of the air temperature and humidity was obtained in a standard United States Weather Bureau shelter, located in the northeast corner of the area.

SURFACE TREATMENTS OF CERTAIN PLOTS IN 1925

Various treatments of the soil surface were under experiment in 1925, but only plots 6, 7, 10, 11 and 15, in which soil temperatures were taken, will be discussed in this paper. The surface treatments of these plots were as follows:

Plot 6—covered with Pabco Thermo-Gen 214, black on both sides and with large, irregularly triangular perforations 1¼ inches apart.

Plot 7—not covered with paper, cultivated 4 inches deep once a month.

Plot 10—covered with Moistite Thermo-Gen mulch paper, gray on both sides and with small triangular perforations ¾ of an inch apart.

Plot 11—covered with Thermo-Gen, black on both sides, but without perforations.

Plot 15—covered with Mulch Paper Plain, gray on both sides and without perforations.

Where the paper mulch was put on, it was laid in 36-inch strips running north and south. A lap of 3 inches was allowed and over this redwood battens were laid and stapled down with No. 6 iron wire.

These battens were 1¼ inches wide and ¼ inch thick (fig. 3). All plots were covered on May 1 and during the 1925 season no crop was grown on any of them.

A period of very high temperatures with clear weather occurred in California, centering around July 16–18, 1925. At some points in the state all previous maximum temperatures for July were equalled or exceeded. The discussion in this paper is confined to a consideration of the temperature changes recorded during this week, or from July 14 to July 21.

Fig. 3.—Paper mulch and bare plots, Davis, California, May, 1925.

EXPERIMENTAL RESULTS

In two plots (Nos. 6 and 7) temperatures were obtained at the depth of one-half inch. The average day temperature (sunrise to sunset) in the bare plot, No. 7, at this depth was 109°, and in plot 6, which was covered with black perforated paper, it was 99°, or 10 degrees colder. The night (sunset to sunrise) temperatures were reversed, however, the average of the bare plot being 81° and of the covered plot 86.6°, or a difference in favor of the covered plot of 5.6°. The highest soil temperature recorded in any of these plots in 1925 was 143° obtained at a depth of one-half inch in the bare plot (No. 7) on July 17.

At the six-inch depth in these same two plots the average day temperature was 91.3° for the bare plot and 90.4° for the covered plot, or a difference of only 0.9° in favor of the bare plot. At the

same depth the average night temperature was slightly higher in both cases, it being 92.7° for the bare plot and 92.1° for the covered plot.

A more detailed study of the records from the bare plot at the various depths shows that the average day and night temperatures (fig. 4) to a depth of twelve inches during this warm period differ decidedly; but at greater depths little or no difference was noted. This is in full agreement with the records for any of the clear days in 1925 not herewith reported, as temperatures were obtained from February to October.

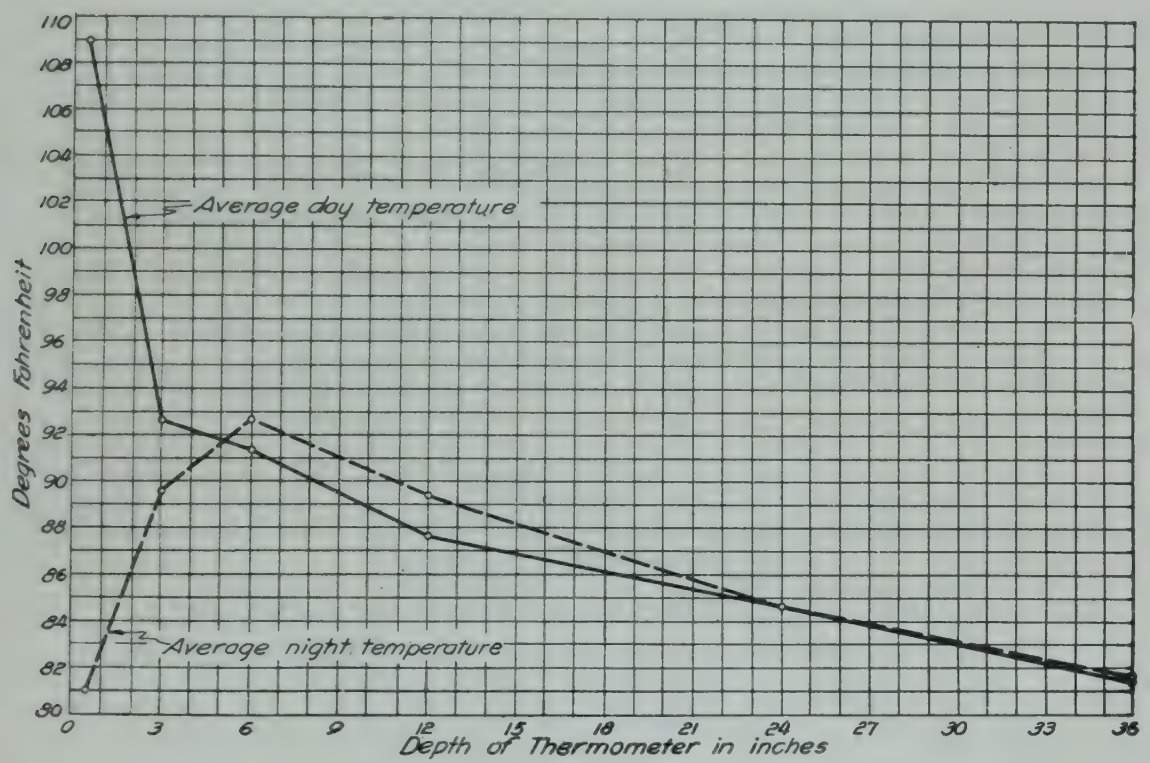

Fig. 4.—Temperature ranges in bare plot, July 14–21, 1925.

The average time of sunrise during the week of July 14–21 was 4:54, and for sunset it was 7:30. The time of occurrence of the maximum and minimum temperatures in the bare plot are shown in table 1. There was no diurnal effect below 12 inches, but there was a gradual increase in temperature, which at the 24-inch depth amounted during the week to four degrees. The maximum air temperature for the week occurred at approximately the same time as that at the one-half inch depth, while the minimum air temperature occurred 28 minutes earlier than the minimum at the one-half inch depth.

The rate at which the heat moved into the soil is shown graphically in figure 5 and the loss of heat from the soil is illustrated in figure 6.

TABLE 1

TIME OF OCCURRENCE OF MAXIMUMS AND MINIMUMS IN BARE PLOT

Depth	Time of maximum after sunrise	Time of minimum after sunset
½ inch	8 hours 51 minutes	8 hours 51 minutes
3 inches	11 hours	10 hours 43 minutes
6 inches	12 hours 17 minutes	11 hours 51 minutes
12 inches	16 hours 25 minutes	14 hours

The gradual increase in temperatures at depths below 12 inches shows that heat was not only lost by radiation from the soil to the atmosphere, but also by conduction downward.

The diurnal range in temperature in the bare plot for depths to 12 inches naturally varies inversely to the depth and is shown in figure 7.

When the temperatures at the 3 inch depth in all five plots are compared (figs. 8, 9), it is quite evident that different results were obtained.

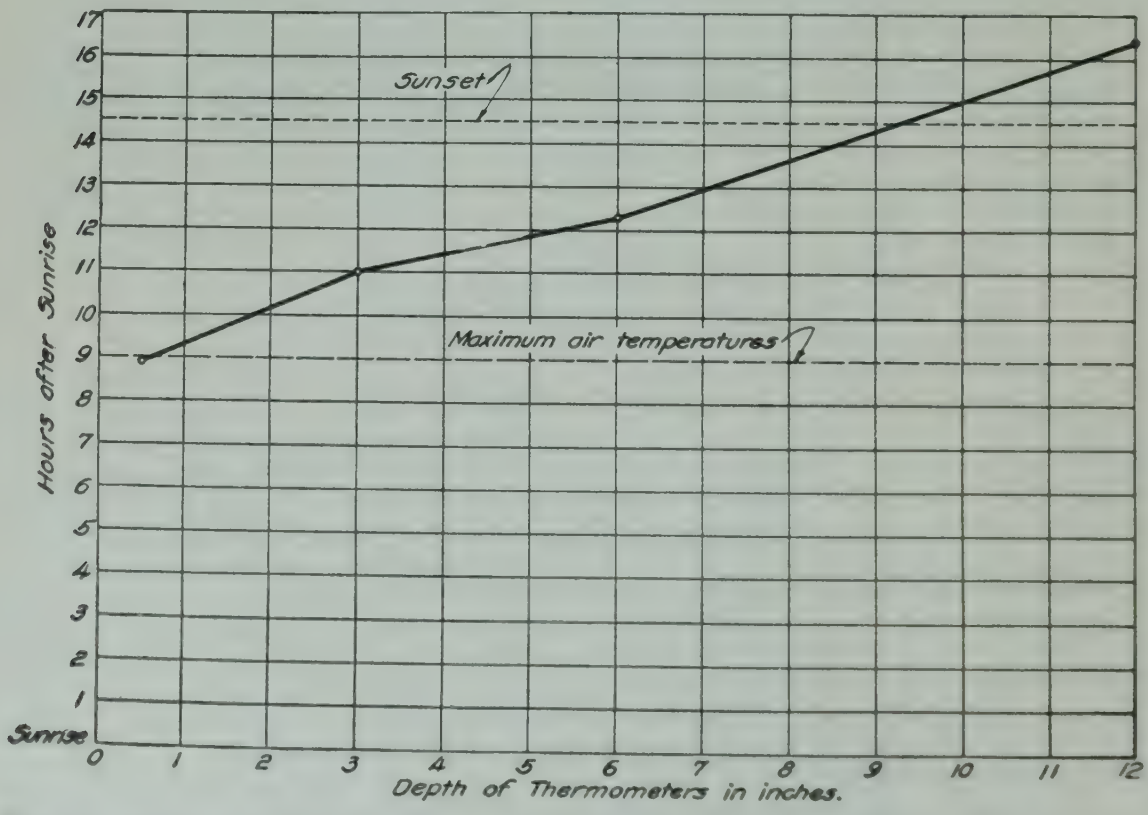

Fig. 5.—Maximum temperatures in bare plot: time of occurrence after sunrise. Average for the week of July 14–21, 1925.

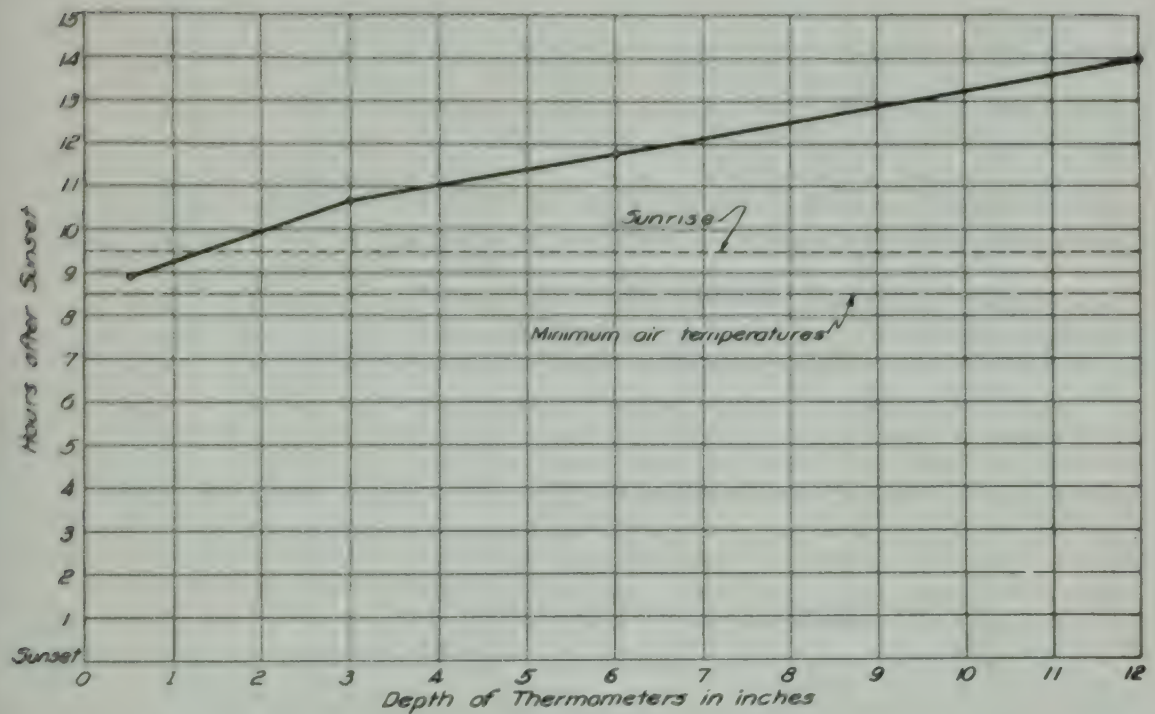

Fig. 6.—Minimum temperatures in bare plot: time of occurrence after sunset. Average for the week of July 14–21, 1925.

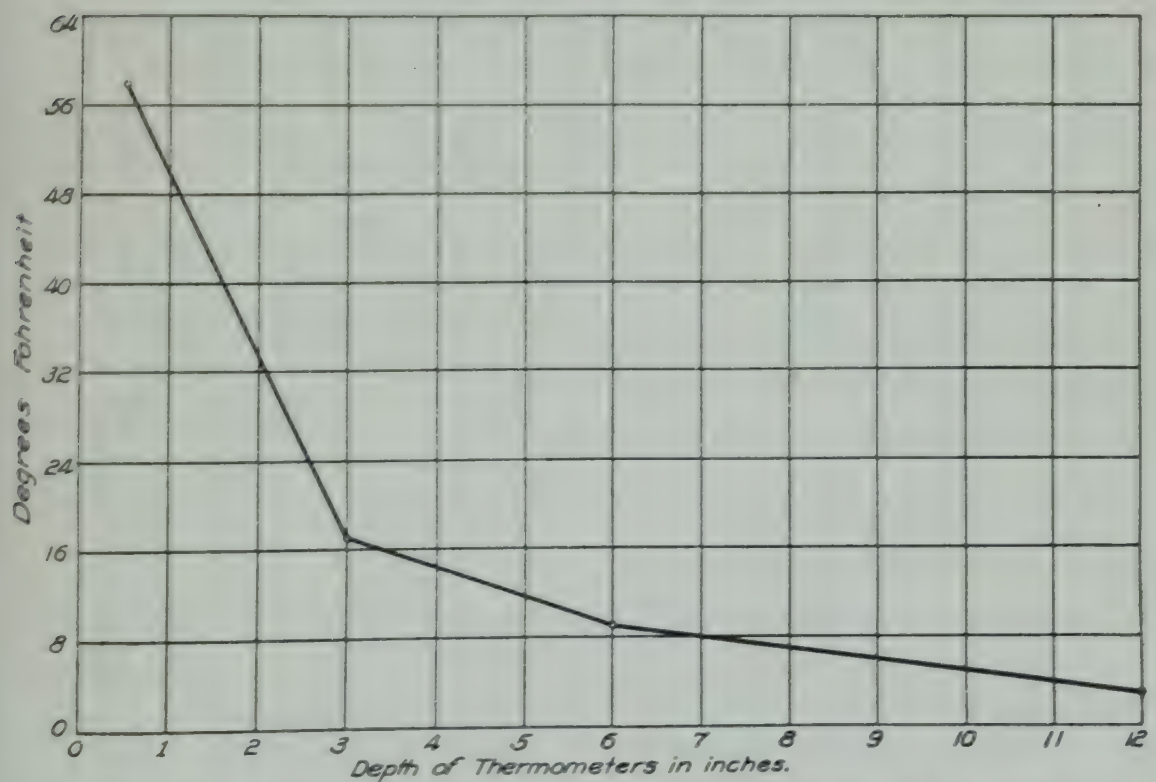

Fig. 7.—Average diurnal range for the week of July 14–21, 1925.

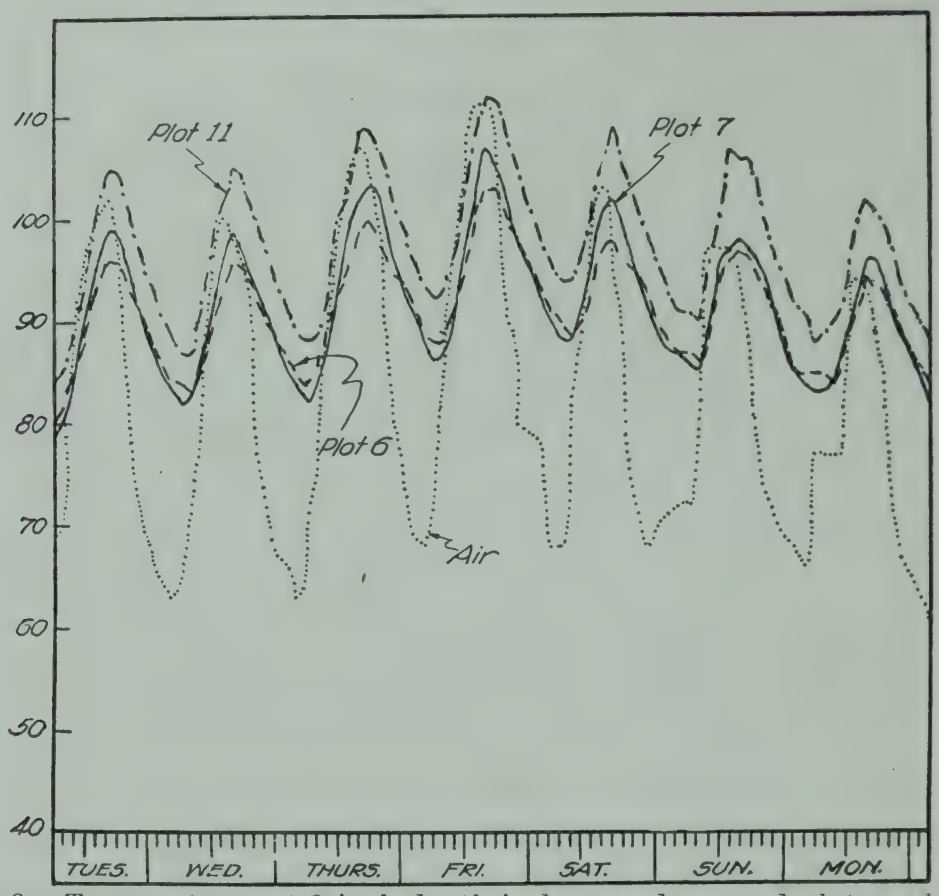

Fig. 8.—Temperatures at 3 inch depth in bare and covered plots and of air by two hour intervals. Week of July 14–21, 1925.

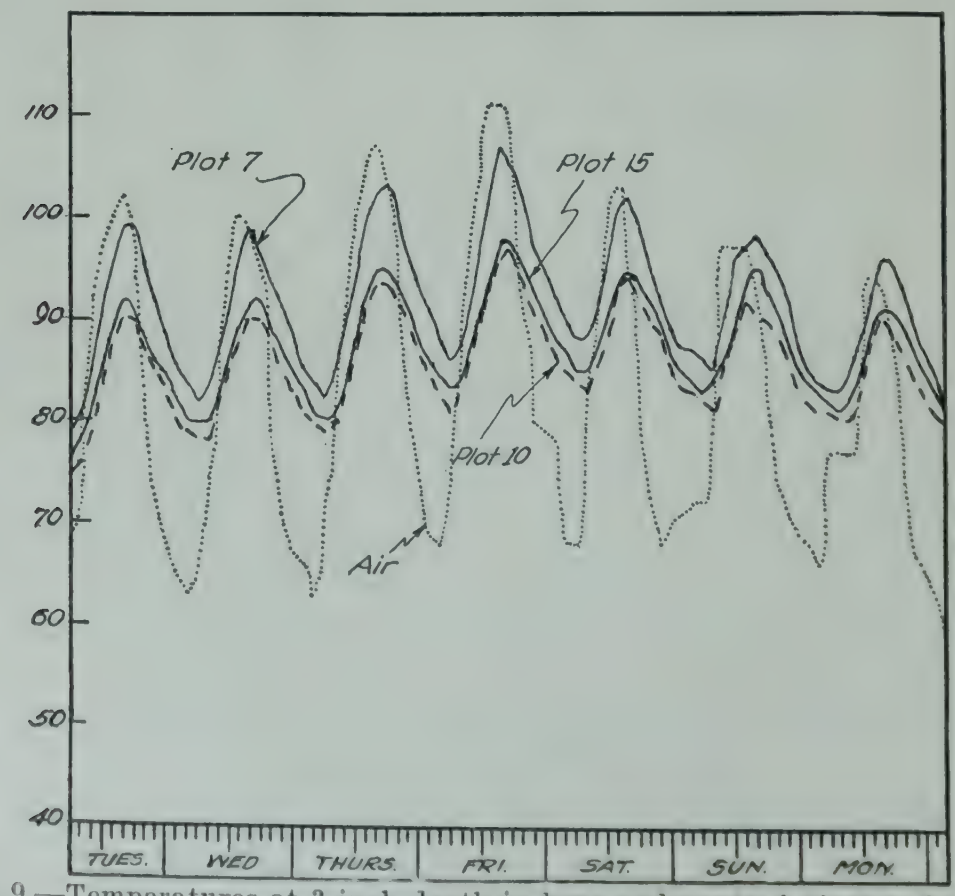

Fig. 9.—Temperatures at 3 inch depth in bare and covered plots and of air by two hour intervals. Week of July 14–21, 1925.

TABLE 2

AVERAGE TEMPERATURES AT THREE-INCH DEPTH, JULY 14–21, 1925

Plot	Cover	Day	Night	Maximum	Minimum
		° *F.*	° *F.*	° *F.*	° *F.*
11	Black solid paper	98.0	96.0	107.0	88.7
7	No paper—soil mulch	92.6	89.6	100.5	83.4
6	Black perforated paper	91.4	90.4	97.9	84.7
10	Gray perforated paper	86.3	85.0	92.4	79.4
15	Gray solid paper	88.0	86.0	94.0	81.3
Air				102.0	66.9

In table 2 the effect of the color of the paper mulch as well as the effect of perforations is shown. Plot 11, which was covered with black solid paper, was 5.4° warmer during the day and 6.4° warmer during the night than the bare plot (No. 7). Where the black perforated paper was used as a mulch (plot 6), the average day and night temperatures were about the same as in the bare plot, with slightly higher night temperatures in favor of the black perforated paper.

Plot 15, which was covered with gray solid paper, was 4.6° colder during the day and 3.6° colder during the night than the bare plot. The coldest of the five plots was number 10, where the average day temperature was 6.3° lower than in the bare plot (No. 7). As the color of the surface soil in these plots is brown, drying to a light brown, the plots covered with the light-colored or gray papers did not have the capacity to absorb as much heat as the bare plot, while those covered with black paper had an increased capacity for heat absorption.

The effect of perforating the mulch paper with large and frequent perforations (fig. 10) is to permit the circulation of air, which results in general in lower temperatures being recorded at the 3 inch depth in the plots covered with perforated paper than in those covered with the non-perforated paper. Plot 6, which was covered with black perforated paper, was on the average 1.2° colder during the day and slightly warmer during the night, as shown in table 2, than the bare plot. The gray perforated paper (plot 10) was the coldest of the five plots, both during the day and at night. Whatever heat is absorbed by the paper mulches during periods of sunlight is rapidly lost from the soil surface by conduction and radiation to the atmosphere when the papers are perforated.

The maximum temperatures in the five plots at the 3-inch depth occurred usually at about the same time, which was two hours after the maximum air temperature was reached. The average maximums

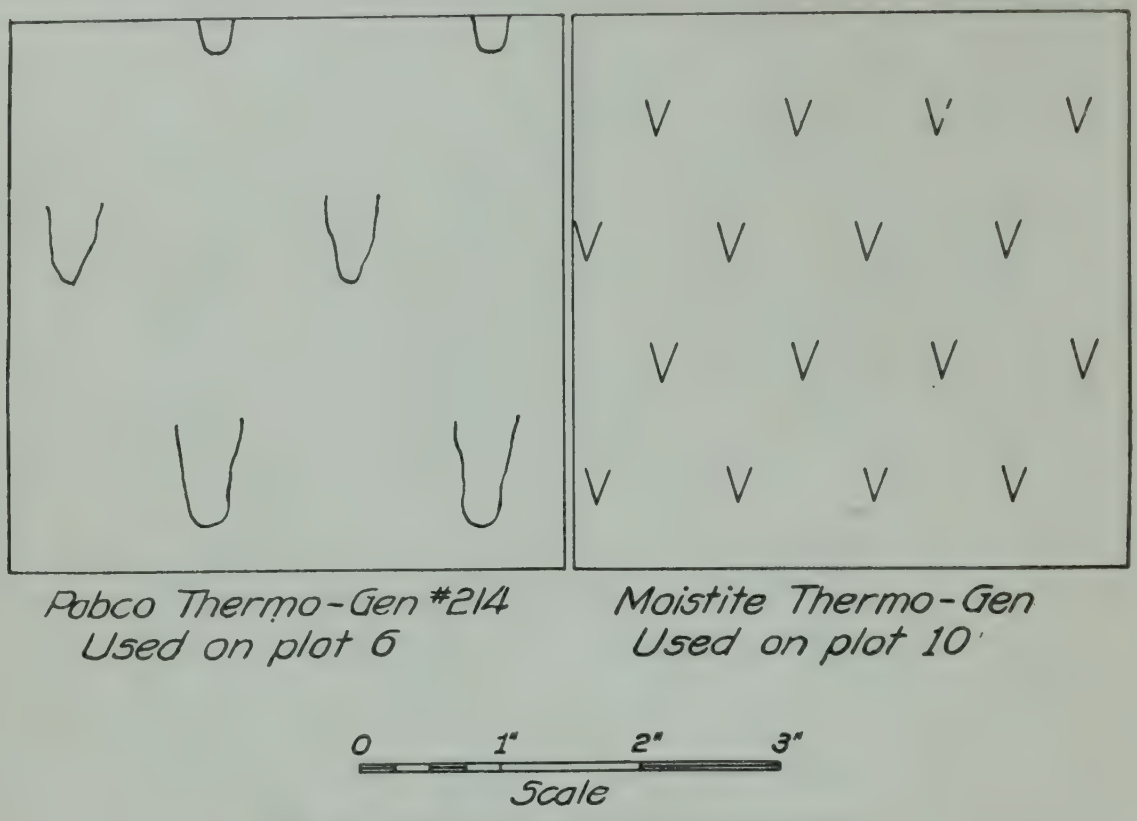

Fig. 10.—Types of perforated paper used.

of these plots as given in table 2 show the range of from 94° to 107°, or 13°. The minimum temperatures at the same depth occurred on the average 1 hour and 40 minutes after the minimum air temperature and showed a range from 81.3° to 88.7°, or 7.4°.

The changes in temperature which occurred at the 12 inch depth in these five plots are graphically shown in figures 11 and 12. The maximum temperatures occurred usually at about the same time, which was 8 hours after the maximum air temperature was reached, and showed a range of 11.6°. The minimums occurred, on the average, 6 hours after the minimum air temperatures and had a range of 10.6°. The results for the 12 inch depth are shown in table 3.

TABLE 3

AVERAGE TEMPERATURES AT TWELVE-INCH DEPTH, JULY 14–21, 1925

Plot	Day	Night	Maximum	Minimum
	° *F.*	° *F.*	° *F.*	° *F.*
11	93.1	95.4	96.0	92.6
7	87.6	89.4	90.3	86.4
6	88.6	90.0	90.6	87.7
10	82.7	84.0	84.4	82.0
15	84.0	85.0	85.6	83.0
Air	.....	.....	102.0	66.9

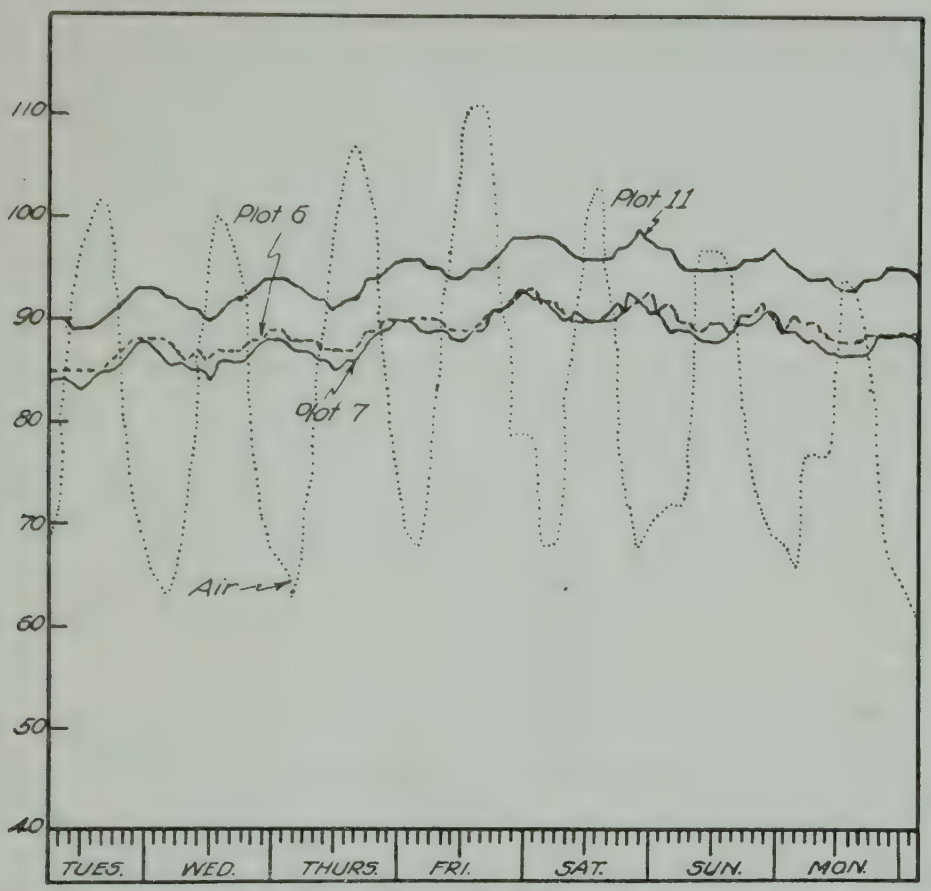

Fig. 11.—Temperatures at 12 inch depth in bare and covered plots and of air by two hour intervals. Week of July 14–21, 1925.

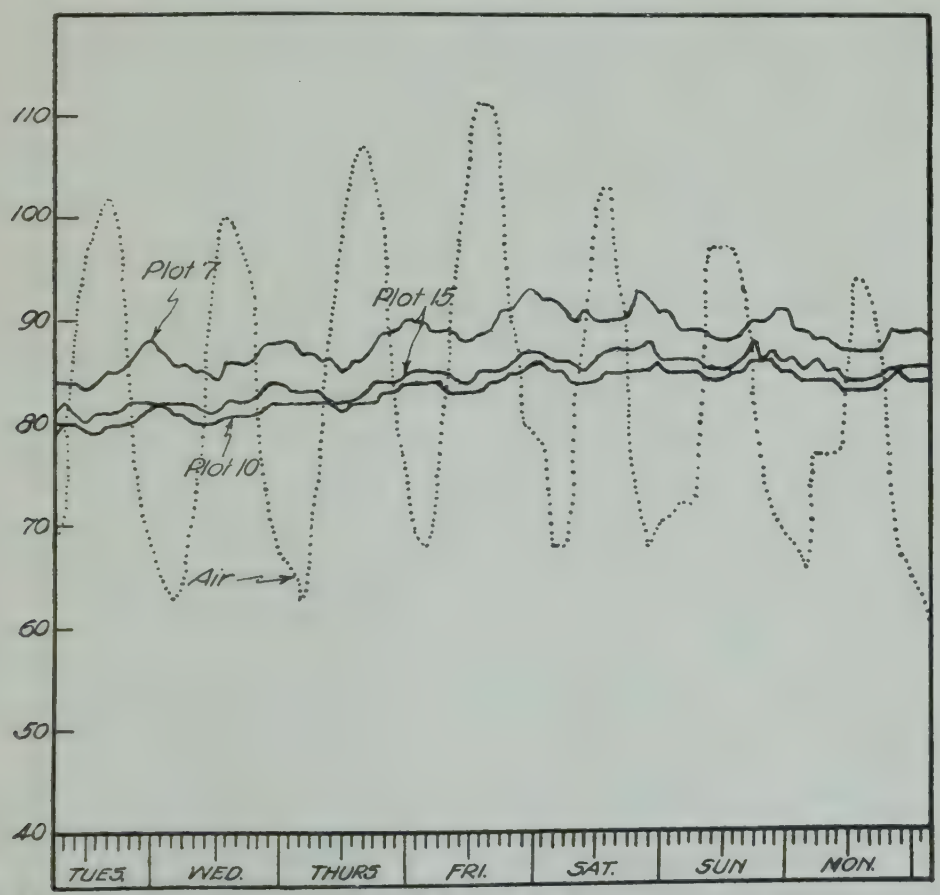

Fig. 12.—Temperatures at 12 inch depth in bare and covered plots and of air by two hour intervals. Week of July 14–21, 1925.

In general the effects of the various types of mulches were the same at a depth of 12 inches (table 3) as those at the 3 inch depth. Plot 11, covered with black solid paper, had the highest temperatures at the 12 inch depth. The average day temperature was 5.5°, and the average night temperature 6.0°, higher than in the bare plot. Plot 6, covered with black perforated paper, was slightly warmer at the 12 inch depth than the bare plot. The plots covered with gray paper mulches were the coldest at the 12 inch depth. Plot 15 covered with gray solid paper was 3.6° and 4.4° colder on the average during the day and night, respectively, than the bare plot (No. 7). Plot 10, covered with the gray perforated paper, was on the average during the day 4.9° and during the night 5.4° colder than the bare plot.

SUMMARY

The highest soil temperature found was in the bare plot, where on July 17, 1925, at a depth of one-half inch, 143° F was registered. Temperatures obtained at a depth of one-half inch showed that the bare plot averaged 10° warmer during the day and 5.6° cooler at night than the plot covered with perforated black paper. In the bare plot the average day temperature at the 6 inch depth for the week was 0.9° higher and the average night temperature was 0.6° higher than on the area covered with perforated black mulch paper.

In the bare plot, where temperatures were obtained at depths of ½, 3, 6, 12, 24, and 36 inches, decided differences were found between the night and day temperatures to a depth of 12 inches.

Temperatures taken at a depth of 3 inches in five plots, where different mulches were used, varied considerably. The warmest plot was that covered with solid black paper. Where the perforated black paper was used the temperatures were about the same as in the bare plot. The coldest plots were those covered with gray paper and in this case the perforated paper was again colder than the non-perforated. The maximum temperatures at the 3 inch depth occurred on the average 2 hours after the maximum air temperature and showed a range of 13°. The minimum temperatures at the 3 inch depth occurred on the average 1 hour and 40 minutes after the minimum air temperature and had a range of 7.4°.

Temperatures taken at a depth of 12 inches showed that the plot covered with the solid black paper was the warmest. The plot covered with perforated black paper was slightly warmer at the 12 inch depth

than the bare plot. The coldest plot was that covered with gray perforated paper, while the gray solid paper was only slightly warmer than the gray perforated. The maximum temperatures at the 12 inch depth occurred usually about 8 hours after the maximum air temperature was reached and showed a range in the various plots of 11.6°. The minimum temperatures at the same depth occurred, on the average, 6 hours after the minimum air temperature and had a range of 10.6°.

The effect of the mulch paper was markedly influenced by variations in color and by the presence or absence of perforations. The warmest soil during this week was that covered with non-perforated black paper, and the coldest that covered with the gray perforated paper. Under the conditions of this experiment, the standard perforated mulch papers showed no material effect in increasing the soil temperatures.

These results are not given as indicating the possible effects which might occur when paper mulches are used in crop rows and are not to be taken as recommendations for the use of any particular type of paper mulch. Other work is in progress which will include the effects on crops. This will be reported later.

LITERATURE CITED

[1] Eckart, Chas. F.

1923. How thermogen enhances the growth of plants 1–29. Published by author, San Francisco.

[2] Hartung, W. J.

1926. The functions of paper mulch in pineapple culture 1–31. Hawaiian Pineapple Co., Ltd., Honolulu.

[3] Lyon, H. L., G. R. Stewart, and Wallace MacFarlane

1923. Univ. Hawaii, second annual short course in pineapple production: 36–39, 74–75, 143–144.

[4] Shaw, C. F.

1926. The effect of a paper mulch on soil temperature. Hilgardia 1: 341–365.

[5] Stewart, G. R., E. C. Thomas, and John Horner

1926. Some effects of mulching paper on Hawaiian soils. Soil Sci. 22: 35–51.

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MAXIMUM HEIGHT OF CAPILLARY RISE STARTING WITH SOIL AT CAPILLARY SATURATION

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INTRODUCTION

The height to which water will be lifted through a soil by film forces, commonly designated as "capillary rise," is an important factor in many phases of agricultural practice, particularly in determining the depth at which the ground water table should be maintained in order to prevent evaporation from the surface.

In most experiments heretofore reported, the capillary rise has been determined by starting with the soil in an air-dry condition and usually in tubes of relatively small diameter.[3] The experiments of Hilgard[1] have generally been quoted to show a maximum rise of 122 inches in the silt separate with less rise in all the other separates, while the work of Linde and Dupre[2] shows that under ideal conditions, where friction of flow through the soil is eliminated, the total height may reach nearly to thirty feet!

Since most soils in agricultural use are frequently or occasionally wetted to the water table by rain or by irrigation, it was felt that to properly measure the maximum possible "capillary rise" under conditions simulating those in the field, the soils should be started at or near capillary saturation, and the ability of the soil to raise water be measured by the amounts removed from a ground-water reservoir and evaporated from the surface.

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FIRST EXPERIMENT

Experimental methods.—In the first experiment, conducted in the laboratories at Berkeley, the soil was placed in galvanized iron tubes 8 inches in diameter, and 4, 6, 8, and 10 feet in length. The soil used was Yolo sandy loam from Davis, California. The soil was placed in the tubes by pouring steadily from the top, gently hitting the sides of the tubes to induce settling. There is no doubt but that there was some stratification of the soil in the tube. The tubes were so arranged that their tops were at the same level, just projecting into a tunnel of muslin (on a frame work) through which a constant stream of warm air was drawn by an electric fan. The air entered the tunnel near large steam pipes, and was generally heated to a temperature of from 70° to 85° F. The bottoms of the tubes were placed in closed reservoirs in which a constant water level was maintained by Winchester supply bottles by which the amount of water taken up by each tube could be measured.

The experiment was set up and irrigation water applied on September 1, 1922. Additional water was applied until drainage occurred, when the water in the reservoir was brought to the predetermined level, the constant supply arranged and the reservoir closed to prevent evaporation losses. As drainage occurred first in the case of the shorter tubes, these had a more extended period of evaporation than the longer tubes. Slow drainage from the soil served to add water to the reservoirs and as this was not removed, the quantity from the 8 and 10-foot tubes exceeded the amount lost by evaporation, giving negative results.

Amounts of water evaporated.—The experimental period was completed and the 4 and 6-foot tubes taken down and sampled on November 27, 1922, after 87 days, and the 8 and 10-foot tubes sampled on December 4 and 5, 1922, after 95 and 96 days. During this time the water was used up rapidly by the 4-foot columns of soil and slowly by the 6-foot columns, while one 8-foot column showed a slight loss and the other 8-foot column and both 10-foot columns showed gains in the water reservoir, due to the excess irrigation water draining from the tube. The loss or gain for each tube is shown in table 1.

The tubes were observed daily throughout the experiment, and while the loss of water from the constant-level replenishment reservoir was noticeable and steady for the 4 and 6-foot tubes, none could be observed from the 10-foot tubes and only a little from one of the 8-foot tubes.

TABLE 1

LOSS OF WATER BY EVAPORATION FROM CAPILLARY RISE TUBES CONTAINING YOLO SANDY LOAM (MOISTURE EQUIVALENT = 16), BERKELEY, 1922

Number of tube	Length	Total water evaporated in liters	Water used daily in grams	Equivalent in surface inches monthly
41	4 feet	3.778	43.4	1.5780
42	4 feet	5.027	57.7	2.1000
61	6 feet	1.295	14.9	.5424
62	6 feet	1.700	19.5	.7098
81	8 feet	.361 (gain)	0	.0000
82	8 feet	.423	4.4	.1599
101	10 feet	.084 (gain)	0	.0000
102	10 feet	.334 (gain)	0	.0000

As the soils were dried by evaporation from the surface, water was drawn by film forces from the deeper wet layers. If these forces could maintain a constant film of water from the reservoir to the surface, continuous evaporation would take place, but if the depth to water table was greater than the film forces could lift the water—greater than capillary rise—then no losses from the reservoir could take place.

Distribution of water.—On taking the tubes down, they were sampled by 1-inch sections by use of a modified King tube, and moisture determinations made. The distribution is shown in table 2.

The rather irregular distribution in places is no doubt due to stratification during filling. The graphs in figure 2 show the distribution when the curves were smoothed.

SECOND EXPERIMENT

Experimental methods.—As the drainage from the longer tubes masked the effects of evaporation and capillary rise, a duplication of this experiment was undertaken at Davis, using care to guard against the errors and difficulties encountered in the original experiment. This work was started in August, 1924, and closed in July, 1925, after a period of over ten months.

Eight-inch galvanized iron tubes were again used, the lengths being the same as before: 4, 6, 8, and 10 feet. The soil was Yolo loam from the Armstrong tract at Davis, a soil heavier in texture than that used in Berkeley. Great care was used in filling the tubes to avoid stratification and to insure even packing and uniform volume weight

TABLE 2

DISTRIBUTION OF MOISTURE IN CAPILLARY RISE TUBES AT THE END OF THE EXPERIMENT. YOLO SANDY LOAM (MOISTURE EQUIVALENT = 16), BERKELEY, 1922

Height above water table *inches*	Per cent of water present (dry basis)							
	41	42	61	62	81	82	101	102
1	23.90	22.99	25.11	25.45	21.90	22.85	22.07	24.55
2	23.93	23.80	25.03	24.19	21.30	23.00	22.70	24.80
3	23.24	24.45	24.70	23.40	20.50	22.18	22.87	25.86
4	24.20	26.25	28.73	24.20	21.30	25.11	22.59	26.39
5	24.95	26.85	16.23	23.20	21.40	21.12	22.80	25.60
6	23.33	27.60	25.79	20.10	22.80	20.24	.....	25.22
7	25.62	27.20	26.10	18.80	22.90	23.40	24.60	25.35
8	24.24	23.99	26.45	20.43	21.70	23.26	24.81	24.08
9	23.28	22.40	26.70	21.18	.....	23.23	24.10	28.30
10	23.21	21.50	25.20	25.50	22.65	30.50	21.55	26.00
11	22.80	19.75	24.90	22.82	22.55	21.18	21.20	25.30
12	22.20	.....	24.00	.....	22.50	20.20	22.65	24.18
13	20.81	19.95	24.90	22.01	18.10?	18.56	21.43	27.35
14	19.15	20.40	24.45	21.60	15.62?	20.80	21.42	22.97
15	19.91	22.57	23.50	19.80	15.50?	19.65	.....	20.78
16	18.77	20.08	22.35	18.26	15.69?	19.41	20.89	18.87
17	19.53	21.70	22.00	18.71	15.76?	16.36	18.44	16.92
18	.....	.....	21.41	18.60	16.59?	18.34	18.56	20.38
19	.....	19.94	17.84	18.40	.....	19.74	19.27	19.52
20	18.51	22.88	19.65	19.50	16.50	19.63	18.43	18.15
21	17.09	20.79	20.05	18.30	17.38	19.00	18.67	17.84
22	18.00	20.30	19.07	17.06	18.22	19.40	17.09	17.70
23	16.21	20.38	19.45	17.13	16.54	19.40	14.95?	17.14
24	16.70	16.27	16.95	17.29	15.56	18.45	20.14?	16.23
25	14.05	17.11	18.43	16.95	14.25	17.71	13.67	16.35
26	12.71	15.82	.....	16.95	15.36	19.65	16.95	13.71
27	12.16	15.27	17.51	16.84	14.50	28.10	15.50	15.21
28	10.57	16.89	16.85	17.99	12.72	15.25	17.52	17.17
29	12.02	16.87	17.15	16.45	13.74	14.36	14.65	17.54
30	.....	.....	17.06	.....	13.40	15.23	14.63	15.22
31	11.52	15.32	16.80	14.60	13.56	15.78	14.12	17.16
32	12.03	15.10	16.90	16.75	14.81	12.48	13.40	15.55
33	11.13	14.45	16.57	16.18	14.71	14.72	15.58	15.02
34	11.11	14.61	15.91	.....	14.24	14.60	13.30?	13.63
35	.....	14.55	15.89	15.42	15.20	15.40	16.40?	14.42
36	.....	14.30	.....	15.35	13.28	13.80	15.38	15.55
37	11.05	13.52	15.25	15.20	13.90	14.92	15.57	14.70
38	10.65	15.05	15.28	15.75	11.95	14.00	14.27	14.50
39	10.60	12.99	16.45	14.39	11.12	14.68	13.27	14.54
40	11.33	13.01	14.89	13.29	12.44	13.39	14.17	14.18
41	9.81	11.09	14.93	14.21	12.94	13.38	14.93	13.46

TABLE 2—(*Continued*)

Height above water table *inches*	Per cent of water present (dry basis)							
	41	42	61	62	81	82	101	102
42	10.12	10.50	15.10	13.91	11.46	14.80	14.03	13.99
43	8.95	8.10	15.04	13.82	12.06	14.00	14.20	12.00
44	8.47	10.05	14.40	13.70	13.34	14.43	13.04	13.20
45	7.58	8.62	16.40	14.08	13.50	14.25	13.21	13.98
46	6.03	6.09	14.94	13.85	11.89	14.33	12.75	13.53
47	3.33	.....	15.78	13.83	12.74	14.31	12.29	11.64
48			15.43	13.60	12.16	13.87	14.81	13.64
49			13.86	13.12	.....	13.31	14.53	13.51
50			14.46	12.15	10.70	14.01	13.92	11.45
51			14.06	11.96	.....	14.00	13.50	13.78
52			.....	12.83	12.05	13.25	12.65	14.52
53			12.90	12.70	12.50	13.96	13.10	13.56
54			13.67	12.20	12.61	10.38	13.43	13.40
55			13.60	12.52	11.20	13.10	13.82	13.10
56			.....	11.53	12.04	11.86	14.78?	12.82
57			13.20	11.89	11.33	13.25	14.00	13.73
58			12.68	13.10	12.26	14.43	13.73	13.07
59			11.75	12.12	12.40	13.04	14.26	13.13
60			11.54	12.11	12.55	13.32	13.51	13.46
61			12.35	14.57	11.75	13.90	13.83	12.60
62			9.15	11.37	12.38	12.46	15.23?	12.32
63			.....	.....	12.90	13.01	13.90	12.56
64			11.20	8.92	11.44	13.28	14.82	12.20
65			10.57	8.31	11.22	13.37	14.39	.....
66			10.10	8.72	11.23	13.92	14.62	12.43
67			10.05	4.64	11.60	13.81	14.23	12.85
68			9.36	6.32	.....	13.50	14.03	12.58
69			5.77	3.56	11.05	13.61	13.60	12.76
70			7.90	.....	10.54	12.70	14.20	.....
71			4.16	.....	11.07	11.85	14.04	11.77
72					10.34	13.50	12.83	15.14
73					11.22	13.44	13.52	20.66
74					11.52	11.71	13.92	12.54
75					.....	12.05	14.45	13.64
76					10.85	11.60	12.81	11.46
77					11.22	12.16	13.58	12.11
78					11.62	12.27	12.51	19.35
79					11.62	12.05	13.21	12.78
80					11.37	11.15	12.84	13.15
81					11.80	11.28	13.59	12.10
82					11.90	10.24	13.21	12.00
83					11.42	13.35	13.02	13.18
84					.....	11.71	14.75	10.94
85					11.61	10.85	11.25	12.26

TABLE 2—(*Continued*)

Height above water table *inches*	Per cent of water present (dry basis)							
	41	42	61	62	81	82	101	102
86					11.05	10.63	11.73	12.26
87					9.92	11.18	12.31	12.67
88					11.22	10.54	12.38	11.95
89					10.52	10.41	12.29	11.33
90					10.52	8.90	12.57	12.01
91					9.12	6.40	11.89	11.71
92					9.22	7.99	12.36	11.67
93					8.65	6.10	11.82	11.13
94					7.40	4.52	12.17	11.38
95					5.36	3.74	12.10	11.80
96					3.30	.....	11.23	12.66
97							12.15	11.70
98							12.35	11.67
99							12.09	10.92
100							11.85	11.38
101							13.95	11.72
102							11.29	11.37
103							12.02	10.95
104							13.93	10.68
105							11.37	11.12
106							12.70	11.11
107							9.12?	11.27
108							12.07	10.85
109							11.23	10.57
110							12.22	10.80
111							10.61	11.23
112							11.91	10.56
113							11.39	10.40
114							9.84	9.55
115							8.82	8.40
116							8.26	7.85
117							7.50	2.26
118							7.32	6.75
119							4.85	4.94
120							2.77	3.30

within the tubes. That this effort was successful is shown by the weight of the soil in the duplicate tubes, and by the volume weight (table 3). The average volume weight was 1.276, with ranges from 1.262 to 1.293, or expressed as pounds weight per cubic foot, an average of 79.66 lbs. with ranges from 78.76 lbs. to 80.73 lbs. A representative sample of this soil had a specific gravity of 2.55, indicating a pore space of almost exactly 50 per cent. The close agree-

ment in the moisture content of the duplicate tubes at the close of the experiment also indicates a uniform packing of the soil.

In this experiment the bottoms of the tubes were set at the same elevation, as shown in figure 1, the tops varying by two-foot intervals. No forced air circulation was attempted, the normal ventilation and circulation of air in the laboratory being relied upon to give comparable evaporation conditions.

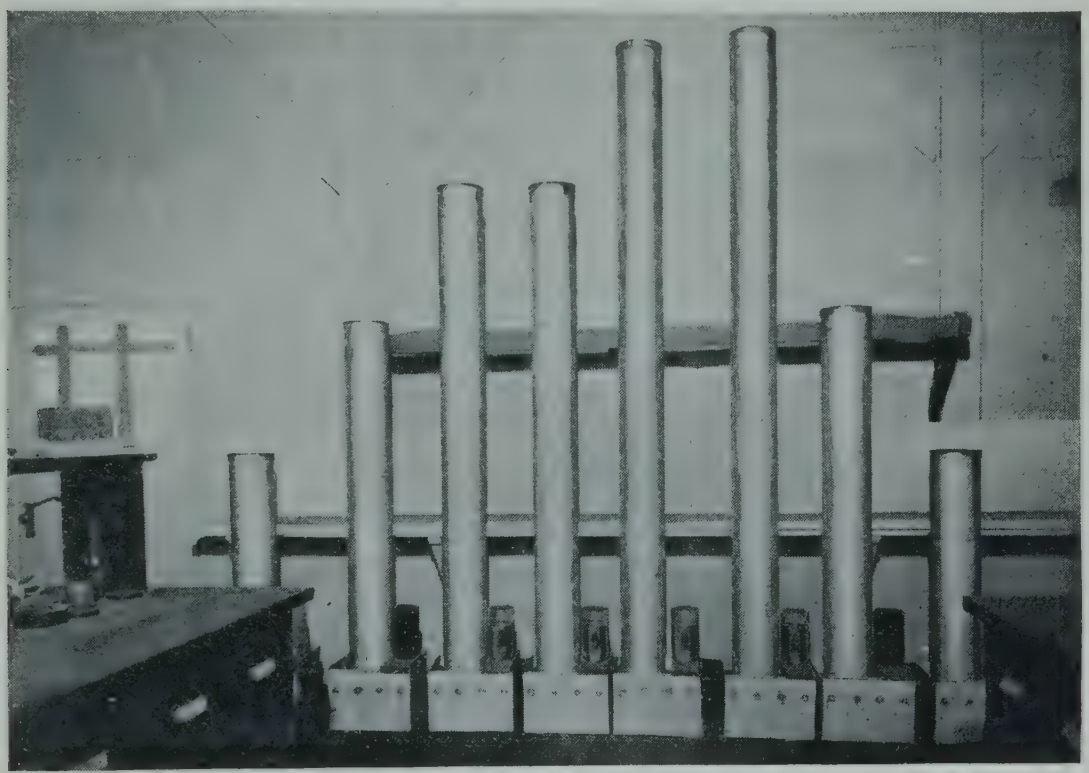

Fig. 1. The eight tubes used in the second experiment, at Davis, showing the reservoirs and the bottles that maintained the constant water level. In the first experiment, at Berkeley, the tubes were so placed that the tops were all at the same elevation and enclosed in a muslin tunnel through which warm air was constantly drawn.

Water was applied to the 10-foot tubes on August 9, to the 6, 8, and 10-foot tubes on August 11, and to all the tubes on August 13, and daily thereafter until August 16. As the soil within the tubes settled, more soil was added to keep them filled to within 1½ inches of the top, and when drainage started, soil was added to completely fill the tubes, a small amount of water being added to wet this soil to the normal moisture condition. Drainage began on August 18 from all except the 10-foot tubes, which began to drain on August 20 and 21. By September 2 drainage from all tubes had apparently ceased.

The water level in the reservoirs was adjusted during the drainage period by removing the excess water and after that period by adding water to the Winchester supply bottles.

Amounts of water evaporated.—The experiment was concluded on July 21, 22, 23, and 24, 1925, when successive tubes were sampled and the distribution of water within the soil columns determined.

TABLE 3

WEIGHT OF SOIL, WATER APPLIED, AND LOSS OF WATER BY EVAPORATION FROM CAPILLARY RISE TUBES CONTAINING YOLO LOAM (MOISTURE EQUIVALENT = 20), DAVIS, 1924–25

Number of tube	41	42	61	62	81	82	101	102
Depth, in inches	48.00	47.00	72.50	72.25	96.50	96.50	119.88	121.00
Average diameter, in inches	8.12	8.07	8.06	8.04	8.12	8.08	8.07	8.06
Kilograms soil	48.120	48.100	72.450	72.080	96.074	97.024	120.054	120.800
Weight per cubic foot, in pounds	78.83	80.46	78.76	80.73	79.26	79.65	80.02	79.62
Volume weight	1.263	1.288	1.262	1.293	1.269	1.276	1.282	1.275
Water applied, in liters	15.850	15.850	22.850	22.850	30.850	30.850	40.850	40.850
Drainage, in liters	.520	.530	1.640	1.410	3.290	3.420	6.280	6.570
Net water retained, in liters	15.330	15.320	21.210	21.440	27.560	27.430	34.570	34 280
Total evaporation, in liters	12.000	12.650	6.350	6.890	3.440	3.550	.550	.600
Period of evaporation, months	10.7	10.8	10.73	10.8	10.73	10.73	10.76	10.8
Period of evaporation, days	321	324	322	324	322	322	323	324
Evaporation per day, in grams	37.38	39.04	19.72	21 26	10.68	11.02	1.70	1.85
Total evaporation in surface inches	37.47	39.92	19.91	21.71	10.42	11.07	1.72	1.88
Evaporation in surface inches monthly	3.50	3.69	1.85	1.99	.97	1.03	.16	.17

The water used, rate of evaporation and other data are given in table 3. The Winchester supply bottles held two liters of water, and only a little over 0.5 liter each was used by the 10-foot tubes. The supply bottles for the 8-foot tubes were renewed on March 9, these tubes using about 3.5 liters each. It was necessary to renew the supply for the 4 and 6-foot tubes at frequent intervals, though the rate of evaporation decreased considerably during the rainy season. The 6-foot tubes used between 6 and 7 liters each, while the 4-foot tubes used over 12 liters.

When the total use of water is expressed as surface inches evaporated monthly, the 4-foot tubes show an average loss of 3.595 inches, the 6-foot tubes an average loss of 1.92 inches, the 8-foot tubes an average loss of 1.0 inch, and the 10-foot tubes an average of only .165 inch. It is felt that ten feet is approximately the maximum height to which this soil can raise water.

Distribution of water.—The distribution of water within these columns was determined by careful sampling by 3-inch sections to a height of 36 inches and by 6-inch sections above that height. The results are given in table 4.

TABLE 4

DISTRIBUTION OF MOISTURE IN SOIL COLUMNS AT END OF EVAPORATION PERIOD.
YOLO LOAM (MOISTURE EQUIVALENT = 20)
(Percentage on oven dry basis)

Distance from base	Tube numbers							
	41	42	61	62	81	82	101	102
inches								
0– 1	35.19	34.99	35.28	35.19	33.71	33.37	33.76	33.58
1– 3	34.13	35.18	34.03	34.56	33.36	33.11	34.06	34.32
3– 6	34.01	35.36	31.01	33.01	34.03	34.21	34.17	34.61
6– 9	31.88	32.96	30.97	31.38	32.50	33.88	33.14	33.16
9– 12	30.45	30.21	29.83	30.22	32.11	30.42	32.81	33.13
12– 15	30.09	28.82	28.09	30.32	31.97	29.19	32.31	31.81
15– 18	27.85	28.92	27.64	28.26	29.83	28.77	30.49	29.12
18– 21	27.68	26.67	26.00	25.75	28.85	28.25	28.64	28.44
21– 24	26.64	27.27	25.80	25.73	27.97	26.38	28.12	27.79
24– 27	24.02	25.13	24.49	24.23	27.47	25.87	27.29	27.73
27– 30	22.74	23.51	23.88	24.19	26.39	25.00	26.64	27.31
30– 33	22.56	22.22	24.00	24.26	24.50	24.38	25.43	26.61
33– 36	21.00	21.73	22.80	23.00	23.99	23.69	25.38	25.41
36– 42	19.62	19.59	22.55	22.51	22.28	23.03	23.66	24.08
42– 48	16.94	16.74	21.82	21.77	22.15	22.22	22.62	22.60
48– 54			20.13	20.88	21.17	21.09	21.22	21.55
54– 60			18.50	19.31	20.64	20.81	21.15	20.47
60– 66			17.16	17.60	20.23	20.71	20.03	20.02
66– 72			13.55	13.62	20.22	20.28	20.01	20.03
72– 78					19.63	19.19	19.19	19.57
78– 84					16.84	17.81	18.51	18.82
84– 90					15.27	16.28	18.46	18.49
90– 96					11.22	11.38	18.23	18.45
96–102							17.96	17.66
102–108							16.14	16.41
108–114							15.05	15.38
114–120							10.20	10.77
Drainage during sampling...	100 cc.	70 cc.	None	40 cc.	50 cc.	50 cc.	40 cc.	30 cc.

It will be noted that the moisture content at the top of the column was greatest in the shorter tube, the average for the 4-foot columns being 16.84 per cent, for the 6-foot columns 13.59 per cent, for the 8-foot columns 11.60 per cent, and for the 10-foot columns only 10.49 per cent. This was not evident in the Berkeley experiment, where the soils were sampled by *one*-inch depths, and the immediate soil surface was air-dry and, in the case of the 4-foot tubes, considerably crusted.

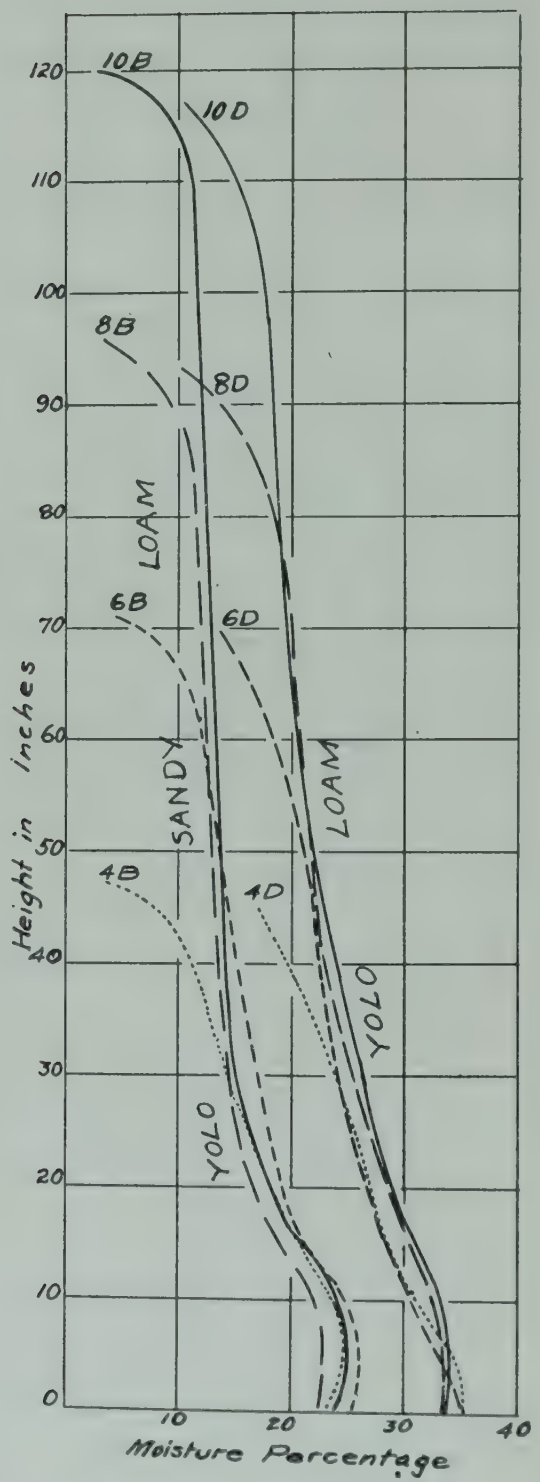

Fig. 2. The distribution of water in the soils at the close of the experiments, after 95 days (B) and 321 days (D) of free evaporation from the surface. (Each curve represents the average of two tubes.) 4B, four-foot tubes at Berkeley; 4D, four-foot tubes at Davis; 6B, six-foot tubes at Berkeley; 6D, six-foot tubes at Davis; 8B, eight-foot tubes at Berkeley; 8D, eight-foot tubes at Davis; 10B, ten-foot tubes at Berkeley; 10D, ten-foot tubes at Davis.

The graphs in figure 2 show the moisture distribution within the tubes from both experiments. The higher water-holding capacity of the Yolo loam as compared to the Yolo sandy loam is shown by the difference of from 8 to 10 per cent of water at any given height. The parallelism of the curves, however, is very striking, although those of the Yolo sandy loam tend to have a steeper slope than those of the loam.

CONCLUSIONS

The Yolo sandy loam and the Yolo loam, wetted to the water table by rains or irrigation, will lift water to the surface at a fairly rapid rate where the water table is within four feet, and at a slower rate if the water table is at six feet below the surface. Some water will be raised to the surface if the water table is at eight feet, but this appears to be close to the limit of such rise, little water being lost from the soil with the water table at ten feet below the surface.

From this it is concluded that with a water table at a depth of more than ten feet below the surface, no losses by evaporation from the surface would occur from a soil having a capillary capacity similar to that of the Yolo sandy loam or Yolo loam. It might be further concluded that for sandy loams and loams in general, water tables at ten feet or more below the surface would be below the maximum height of capillary rise and would result in no movement of water to the surface.

LITERATURE CITED

[1] Hilgard, E. W.
1912. Soils, pp. 188–215. The Macmillan Company, New York.

[2] Linde, C. J., and H. A. Dupre.
1913. On a new method of measuring the capillary lift in soils. Jour. Agron. 5:107–116.

[3] Wadsworth, H. A., and A. Smith.
1926. Some observation upon the effect of the size of the container upon the capillary rise of water through soil columns. Soil Science 22:199–211.

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ACUTE INFECTION OF CHICKS AND CHRONIC INFECTION OF THE OVARIES OF HENS CAUSED BY THE FOWL-TYPHOID ORGANISM*

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INTRODUCTION

The earliest authentic descriptions of fowl typhoid are those of Klein[1] in 1889, in England, and of Moore[2] in 1895–96, in the United States. Klein designated the disease as infectious enteritis and the causative organism, *Bacillus gallinarum*. Moore called the disease infectious leukemia of fowls and the causative organism *Bacterium sanguinarium*.§ It has since been determined that these investigators studied the same disease, which is now known as fowl typhoid and has become recognized as an important cause of mortality of adult fowls throughout the world.

* A brief résumé of these studies was contained in the California Agr. Exp. Sta. Ann. Rpt. 1924–25: 72.

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§ In Bergey's Manual of Determinative Bacteriology (Williams and Wilkins Company, Baltimore, Maryland, 1923), *Bacterium sanguinarium* (Moore) is classified as *Eberthella sanguinaria* (Moore) and is said to be associated with cholera in chickens. The species of organisms stated to be associated with fowl typhoid are *Eberthella jeffersonii* (Hadley), *Eberthella pfaffi* (Hadley), and *Eberthella rettgeri* (Hadley). Klein's *Bacillus gallinarum* is classified as *Pasteurella avicida* (Perroncito), the cause of fowl cholera. This classification is apparently based largely upon the studies of Hadley reported in Rhode Island Agr. Exp. Sta. Bul. 174. This publication, however, presents *E. jeffersonii*, *E. pfaffi*, and *E. rettgeri* as new bacterial types, differing from both the fowl-cholera and fowl typhoid organisms. *Bact. sanguinarium* (Moore) and *B. gallinarum* (Klein) are regarded as identical and as the etiological agent of fowl typhoid. It would appear, therefore, that the fowl-typhoid organism is improperly classified in the above mentioned manual. For this reason *Bact. sanguinarium* (Moore), the name of the fowl-typhoid organism in common usage in the United States, is used in this paper.

Some investigators observed a marked similarity between *Bact. sanguinarium* and *Bact. pullorum*,* the cause of bacillary white diarrhea of chicks. As a result, extensive comparative studies of the two species were made especially by Taylor,[3] Smith and Tenbreeck,[4] Rettger and Koser[5] Goldberg,[6] and Hadley.[7] These investigators concluded that the two organisms were indistinguishable in their serologic reactions but that there were sufficient differences in their action on carbohydrate media, in their other cultural characteristics, and in their morphology to establish the identity of the two species.

References in the literature to fowl typhoid are numerous. The disease, however, has nearly always been described as an acute infection of mature fowls, little consideration being given to the rôle that *Bact. sanguinarium* might play in causing mortality among young chicks.

The organism in recent years has been recovered from dead chicks by several investigators and it therefore can no longer be regarded of importance only in connection with fowl typhoid of adults. Panisset and Verge[8] in 1924 reported an epizootic among a small flock of chicks in France in which they isolated an organism closely resembling *Bact. sanguinarium*. Stafseth in Michigan, Bushnell in Kansas, and Beaudette in New Jersey have stated in personal communications that they have occasionally isolated *Bact. sanguinarium* from chicks which they suspected had died from bacillary white diarrhea. Beaudette[9] in 1925 reported the isolation of the organism both from young chicks and the ovary of a hen and he stated that 9 of 66 hens in the same flock reacted to an agglutination test with *Bact. pullorum* or *Bact. sanguinarium* antigens. In 1926, Doyle[10] reported similar observations regarding the occurrence of the infection in chicks and hens. He also stated that the examination of 140 eggs from 9 reacting hens showed them to be free from *Bact. sanguinarium*. Disease of baby chicks due to *Bact. sanguinarium* was first observed in this laboratory in May, 1921, in chicks submitted for diagnosis. Clinically and in all other respects the disease resembled bacillary white diarrhea. Since then the organism has been occasionally encountered in routine bacteriological examinations of chicks.

In November, 1924, opportunity was afforded to make a detailed study of an outbreak of disease due to *Bact. sanguinarium* in a lot of chicks which had been obtained for experimental purposes. The conditions under which the outbreak occurred made it seem possible that the infection was acquired by a transmission cycle identical with that of *Bact. pullorum*. Investigations were undertaken to determine whether this suspicion was well founded.

* Classified as *Salmonella pullora* in Bergey's Manual of Determinative Bacteriology, p. 218.

THE OUTBREAK OF DISEASE IN BABY CHICKS

On November 14, 1924, 145 chicks were obtained from a commercial hatchery for use in a coccidiosis control experiment. These chicks, all of which appeared vigorous on arrival, had been shipped in new boxes immediately upon removal from the incubator and were about thirty-six hours old when received. They were placed in pens which not only had been thoroughly disinfected but also had not previously contained poultry. The electric hovers, mash hoppers, and drinking fountains used were new and had also been disinfected. These chicks, therefore, were not exposed to infection of any kind except that which might have been present in the incubator or within or on the shell of the egg from which they were hatched.

The day following that on which the chicks were received, when they were about 60 hours old, the death of one chick occurred. *Bact. sanguinarium* was isolated in pure culture. Deaths from this cause continued and became so numerous that the coccidiosis control experiment for which the chicks were secured was abandoned. The outbreak of the disease due to *Bact. sanguinarium,* however, proved of equal interest and these chicks were, therefore, held for study and observation.

TABLE 1

RECORD OF MORTALITY AND RESULTS OF POST-MORTEM EXAMINATION OF ONE HUNDRED AND FORTY-FIVE CHICKS RECEIVED NOVEMBER 14, 1924

Time of death	Number died	Per cent died	Abnormal liver*		Unabsorbed yolk		*Bacterium sanguinarium* isolated		Bacteriological examination negative	
			Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent
First week	29	20.0	29	100.0	28	96.5	27	93.1	2	6.9
Second week	23	15.8	22	95.6	14	60.8	22	95.6	1	4.3
Third week	6	4.1	2	33.3	3	50.0	4	66.6	2	33.3
After third week	21	14.5	4	19.0	2	9.5	4	19.0	17	80.9
Total	79	54.4	57	72.1	47	59.5	57	72.1	22	27.8

* Abnormalities consisted of uniformly yellowish or mottled yellow and red discoloration or uniformly pale without any marked discoloration.

A careful autopsy and bacteriological examination was made of each chick that died. Microscopic, biochemic, and serologic methods were used for identification of cultures. Those that consisted of Gram-negative non-motile rods; that produced acid but no gas in dextrose,

mannite and maltose broth and did not ferment lactose and saccharose broth; and that were agglutinated by positive *Bact. pullorum* and *Bact. sanguinarium* serum but not by positive *B. avisepticus* serum were considered to be *Bact. sanguinarium.*

Table 1 gives a record of the mortality and results of post-mortem examination of the chicks which died before they were 45 days old.

DISCUSSION OF BABY CHICK MORTALITY

The mortality in this lot of 145 chicks during the first 45 days of their lives was 79, or 54.4 per cent. *Bact. sanguinarium* was isolated from 57, or 72.1 per cent, of those that died, or 39.3 per cent of the total number of chicks.

Forty-nine, or 85.9 per cent, of the deaths from fowl-typhoid infection occurred during the first two weeks. Failure to recover the organism was encountered in only 3 of the 52 chicks which died during this period. Of the 27 chicks which died after the second week, *Bact. sanguinarium* was recovered from 8, or 29.6 per cent.

The distribution of abnormal livers and unabsorbed yolks with respect to the age of the chicks at the time of death corresponded closely to the incidence of *Bact. sanguinarium* infection. Abnormalities of the liver were found in 51, or 98.0 per cent of the 52 which died during the first two weeks and in 6, or 22.2 per cent, of those which died later. Unabsorbed yolk was present in 42, or 80.7 per cent, of those which died during the first two weeks and in 5, or 18.5 per cent, of those which died later.

It can be seen that this outbreak of disease due to *Bact. sanguinarium* resembled in all respects bacillary white diarrhea of baby chicks due to *Bact. pullorum* infection.

OBSERVATIONS ON THE SURVIVORS

Twenty-five of the survivors, 20 females and 5 males, were kept for further study. An agglutination test with *Bact. sanguinarium* antigen was made on the blood serum of each bird when they were six, eight, and twelve months of age. No reactions occurred. One bird died when seven months old. No pathological changes were found in the ovary and a bacteriological examination was negative. One bird died when ten months of age. The port-mortem examination showed three small abnormal-appearing yolks in the ovary. The bacteriological examination of the liver, heart blood, and the three

yolks was negative. Since negative results were obtained from the three agglutination tests and no evidence of *Bact. sanguinarium* infection was found in the two birds which died, no further tests or examinations were made of the remaining birds.

While these studies failed to demonstrate that the survivors of an outbreak of fowl typhoid in baby chicks become chronic carriers of *Bact. sanguinarium* they do not preclude the possibility that some did become carriers, since only 25 of the 66 survivors were retained for observation.

INFECTION OF THE OVARIES OF HENS

As stated at the outset, the only possible sources of the infection in the chicks seemed to be either the incubators in which or the eggs from which they were hatched. Since the nursery trays and nursery tray cloths of the incubators were cleaned between hatches and little complaint of chick mortality had been experienced by the hatchery owner, the eggs seemed the most likely source of the infection. This suggested that chronic ovarian infection with *Bact. sanguinarium* might exist in breeding fowls and be transmitted to chicks through the egg in the same manner as *Bact. pullorum*. If this should prove to be true, it seemed possible that the carriers of the infection might be detected by means of the agglutination test. Permission was obtained to collect blood samples for the agglutination test from a portion of the flock of 1300 birds that produced the eggs from which the chicks were hatched.

On December 12, 1924, blood was drawn from 196 of the 1300 birds. The agglutination test was made on each blood sample with antigens prepared from both *Bact. pullorum* and *Bact. sanguinarium*. Positive reactions were obtained with 32 or 16.3 per cent.

The degree of agglutinations obtained with the positive sera is given in table 2.

An analysis of table 2 shows:

1. Partial or complete agglutination with both antigens was obtained with 29 of the 32 samples.

2. Complete agglutination with both antigens in at least one dilution was obtained with 22 samples.

3. In three instances (Nos. 444, 461, 479) there was complete agglutination with *Bact. pullorum* antigen but only partial with *Bact. sanguinarium* antigen. Repetition of the tests with these samples gave the same results.

4. One sample (No. 416) gave complete agglutination with *Bact. sanguinarium* antigen but only partial agglutination with *Bact. pullorum* antigen. Repetition of the test with this sample gave the same result.

TABLE 2

THE REACTIONS TO THE AGGLUTINATION TEST OF THE POSITIVE SERA

Bird No.	*Bact. pullorum* antigen		*Bact. sanguinarium* antigen		Bird No.	*Bact. pullorum* antigen		*Bact. sanguinarium* antigen	
	0.02 mil serum	0.01 mil serum	0.02 mil serum	0.01 mil serum		0.02 mil serum	0.01 mil serum	0.02 mil serum	0.01 mil serum
304	+	∓	+	−	395	+	+	+	+
315	+	±	+	±	415	+	−	+	−
320	±	+	±	±	416	∓	−	+	−
325	±	−	∓	−	423	+	∓	−	−
341	±	±	±	±	425	±	+	±	±
349	+	−	−	−	432	∓	∓	±	−
350	+	−	−	−	435	+	∓	+	∓
354	±	±	±	±	443	+	+	+	±
365	+	+	±	±	444	±	+	±	−
368	±	+	±	+	452	±	±	±	±
371	±	±	±	±	456	±	±	±	±
373	+	−	±	±	461	+	±	∓	−
381	±	+	±	±	464	+	+	±	±
382	±	±	±	+	465	+	∓	+	∓
383	+	−	+	+	477	∓	∓	∓	−
394	±	+	+	+	479	+	+	∓	∓

Explanation of symbols in table:

Two readings were made of each test at the expiration of 24 and 48 hours, respectively. Only final readings were entered.

Two dilutions were made by adding 0.02 mil and 0.01 mil of serum respectively, to 1 mil of antigen.

− indicates no evidence of reaction.

∓ indicates slight sediment but supernatant fluid turbid.

± indicates more sediment than ∓ but still a faint cloudiness in supernatant fluid.

± indicates that after over-night incubation complete agglutination is present.

+ indicates that after over-night incubation there was not complete agglutination, but on standing for 24 hours longer the fluid cleared up.

5. Partial agglutination with both *Bact. pullorum* and *Bact. sanguinarium* antigens was obtained with three samples (Nos. 325, 432, 477). Repetition of the tests with these samples gave the same results.

6. Complete agglutination with *Bact. pullorum* antigen but *none* with *Bact sanguinarium* antigen was obtained with three samples (Nos. 349, 350, 423). Repetition of the tests with these samples gave the same results.

The results of the agglutination test indicated that the reacting birds were infected either with both *Bact. pullorum* and *Bact. sanguinarium* or with one of the two species alone, but did not make it possible to arrive at a more definite conclusion. In order that information on this point might be obtained 29 of the 32 reactors were secured

for autopsy. The three reactors not obtained were numbers 315, 435, and 452, all of which had reacted positively with both antigens. The birds obtained were examined for abnormalities, particularly of the ovary. Cultures were made from the livers, spleens, ovaries, and normal and abnormal-appearing ovules and yolks. The results of the autopsies and bacteriologic examinations appear in table 3.

DISCUSSION OF POST-MORTEM AND BACTERIOLOGICAL FINDINGS

Abnormal ovaries were found in all of the twenty-nine reacting hens examined. The abnormalities of the ovaries included bloody or caseated ovules; yolks varying from a pea to a hazelnut in size with a thick opaque capsule containing yellow semi-solid oily material or a clear yellow oily liquid with white flakes in suspension; small partially solidified, blood-tinged yolks; yolks of various sizes with capsule but partially filled with a thick yellow or greenish-yellow liquid; solidified angular yellow or greenish-yellow or blood-tinged yolks; and a number of small cysts attached to the ovary.

No correlation was found to exist between the degree of reaction to agglutination test of the blood serum of the birds and the extent of ovarian abnormalities present. For example, serum from bird 395 in which only slight ovarian abnormalities were found, caused complete agglutination in both dilutions with both antigens, while serum from bird 416, in which extensive abnormalities of the ovaries were found, produced partial agglutination in the 1–50 dilution with the *pullorum* antigen, complete agglutination in the 1–50 dilution with the *sanguinarium* antigen, but no agglutination in the 1–100 dilution with either antigen.

In addition to abnormalities of the ovaries, in seven birds yolk material was found in the abdominal cavity. In three of these (365, 416, 456) the material had the appearance of having escaped from a recently ruptured yolk. In the other four cases (341, 350, 383, 423) the material was solidified or encapsulated indicating that it had been in the abdominal cavity for some time. In two other birds (354, 464) there was no free yolk material in the abdominal cavity but the peritoneum was thickened and opaque suggesting that rupture of a yolk had previously occurred in these birds. The owner stated that a number of the flock had died from ruptured yolk during the preceding laying season. None of the latter were given a bacteriological examination so it is not known that *Bact. sanguinarium* was associated with

TABLE 3

Results of Examinations of Reacting Hens

Bird No.	Agglutination test reaction		Condition of ovary	Description of abnormal yolks	Culture made from	Growth obtained from	Organism recovered
	Bact. pullorum	*Bact. sanguinarium*					
304	+∓	+−	Dormant	1 very bloody ovule	Liver, spleen, 1 normal ovule, 1 bloody ovule, ovary	None	None
320	∓+	∓±	Dormant	1 small yolk, capsule thick and opaque, contents yellow and semi-solid	Liver, spleen, 1 abnormal yolk	Abnormal yolk	*Bact. sanguinarium*
325	±−	∓−	Dormant	2 small semi-solid, blood-tinged yolks	Liver, spleen, 2 abnormal yolks	2 abnormal yolks	*Bact. sanguinarium*
341	∓∓	∓∓	Dormant	3 encapsulated masses of yolk in abdominal cavity. 2 yolks with thick opaque capsules containing clear yellow oily liquid. 1 with capsule not filled, contents greyish-yellow in color. Several other small yolks	Liver, spleen, 3 encysted abdominal yolks, 2 abnormal yolks	Abdominal yolks, abnormal yolks	*Bact. sanguinarium*
349	+−	−−	Dormant	Several small yolks yellowish green in color, contents liquid, capsule not filled	Liver, spleen, 4 abnormal yolks	2 abnormal yolks	*Bact. sanguinarium*
350	+−	−−	Active	1 encapsulated mass of yolk size of a hazelnut in abdominal cavity, contents greyish-yellow thick liquid, numerous minute caseated ovules. Several pea-sized abnormal yolks, capsules not filled, contents yellow liquid	Liver, spleen, 4 caseated ovules, egg in abdominal cavity, 2 abnormal yolks, 2 normal yolks	2 abnormal yolks	*Bact. sanguinarium*
354	∓∓	∓∓	Dormant	1 solidified, angular, and blood-tinged yolk. 1 large, bloody yolk, capsule not filled, contents liquid. 2 small yolks, contents clear, yellow oily liquid, containing white flakes, capsule opaque	Liver, spleen, 4 abnormal yolks	2 abnormal yolks	*Bact. sanguinarium*

TABLE 3—(*Continued*)

Bird No.	Agglutination test reaction		Condition of ovary	Description of abnormal yolks	Culture made from	Growth obtained from	Organism recovered
	Bact. pullorum	*Bact. sanguinarium*					
365	++	±±	Active	Free liquid yolk material in abdominal cavity. 2 abnormal yolks, capsules not filled, yellow liquid contents. 2 small angular solidified yolks	Liver, spleen, abdominal yolk, 4 abnormal yolks	Abdominal yolk, solidified yolks	*Bact. sanguinarium*
368	±+	±+	Dormant	1 entire egg in abdominal cavity, contents creamy, foul odor. Numerous normal appearing ovules	Liver, spleen, abdominal egg, ovules, ovarian tissue	Egg, ovarian tissue, ovules	*Staphlococcus* from egg. *Bact. sanguinarium* from ovary and ovule
371	±±	±±	Active	1 solidified yolk. Several small yolks, capsules not filled, contents liquid	Liver, spleen, 3 abnormal yolks, 1 normal yolk	1 abnormal yolk	*Bact. sanguinarium*
373	+−	±±	Dormant	2 yolks, hazelnut size; capsules thick and opaque, contents clear, yellow, oily liquid with white flakes, 2 yolks, capsules not filled; contents yellow liquid. 1 yolk, capsule not filled, contents bloody	Liver, spleen, 5 abnormal yolks	All 5 yolks	*Bact. sanguinarium*
381	±+	±±	Dormant	Several yolks size of large pea, contents solidified and blood-tinged; 1 slightly larger, contents greyish yellow color and viscid	Liver, spleen, 3 solidified yolks, 1 liquid yolk	3 solidified yolks	*Bact. sanguinarium*
382	±±	±+	Dormant	One half-size yolk, greenish brown, liquid contents, capsule not filled. Several caseated ovules, 3 to 4 mm. in diameter. 2 small semi-solid yolks, capsules not filled, greyish brown in color	Liver, spleen, large yolk, 2 small yolks, ovules	2 small yolks	*Bact. sanguinarium*
383	+−	++	Dormant	1 large and several small solidified angular yolks. Caseated material in abdominal cavity, probably from ruptured yolk. Peritonitis	Liver, spleen, yolk in abdominal cavity, large and small abnormal yolks	Large and small abnormal yolks	*Bact. sanguinarium*

TABLE 3—(*Continued*)

Bird No.	Agglutination test reaction		Condition of ovary	Description of abnormal yolks	Culture made from	Growth obtained from	Organism recovered
	Bact. pullorum	*Bact. sanguinarium*					
394	±+	++	Active	Numerous small cysts in ovary	Liver, spleen, ovary, ovarian cysts	Cysts	*Bact. sanguinarium*
395	++	++	Active	2 small yolks, capsules not filled	Liver, spleen, 2 abnormal yolks, 1 normal yolk	None	None
415	+−	+−	Dormant	2 small yolks, capsules thick and opaque, contents semi-solid and bloody	Liver, spleen, 2 abnormal yolks	Abnormal yolks	*Bact. sanguinarium*
416	∓−	+−	Active	Blood-tinged liquid yolk in abdominal cavity. Several pea-sized abnormal yolks, contents clear yellow oily liquid with white flakes. 1 large abnormal yolk, capsule not filled, contents yellow liquid	Liver, spleen, abdominal yolk, 2 small and 1 large abnormal yolks, 1 normal yolk	1 abnormal yolk	*Bact. sanguinarium*
423	+∓	−−	Dormant	Small encysted mass of solidified bloody yolk in abdomen. One abnormal yolk, capsule not filled, contents yellow thick liquid	Liver, spleen, abdominal yolk, abnormal yolk	Abnormal yolk	*Bact. sanguinarium*
425	±+	±±	Active	Three half-size abnormal yolks, capsules thickened, contents partially solidified. 1 very bloody, others yellow in color	Liver, spleen, normal yolk, 3 abnormal yolks	2 abnormal yolks	*Bact. sanguinarium*
432	∓∓	±−	Dormant	1 hazelnut-sized abnormal yolk, capsule not filled, contents yellow liquid. Several small abnormal yolks of same character	Liver, spleen, 2 abnormal yolks	Both abnormal yolks	*Bact. sanguinarium*
443	++	+±	Active	1 yolk 1 cm. diameter, capsule thick opaque, contents yellow, solidified, 1 yolk 5 mm. in diameter, capsule not filled, contents semi-solid and yellow	Liver, spleen, abnormal yolks, normal yolks	Abnormal yolks	*Bact. sanguinarium*

TABLE 3—(*Concluded*)

Bird No.	Agglutination test reaction		Condition of ovary	Description of abnormal yolks	Culture made from	Growth obtained from	Organism recovered
	Bact. pullorum	*Bact. sanguinarium*					
444	∓+	±−	Active	1 large yolk, capsule not filled, contents thick yellow liquid. 2 large yolks, capsules not filled, contents thick blood-tinged liquid. 1 hazelnut size yolk, capsule thick, opaque, contents clear yellow, oily liquid with white flakes	Liver, spleen, 4 abnormal yolks, 1 normal yolk	4 abnormal yolks	*Bact. sanguinarium*
456	∓∓	∓∓	Active	Liquid yolk in abdominal cavity. 1 yolk, capsule not filled, contents thick yellowish brown liquid. 1 yolk solidified, angular, yellowish brown. 3 yolks, capsules thick, opaque, contents clear yellow oily liquid with white flakes	Liver, spleen, abdominal yolk, abnormal yolks	All abnormal yolks	*Bact. sanguinarium*
461	+±	∓−	Active	1 large, solidified angular bloody yolk. 2 yolks hazelnut-size, capsules thick, opaque, contents solidified, yellow in color. 1 hazelnut-size, capsule thick, opaque, contents yellow, oily liquid with white flakes. Numerous small abnormal ovules	Liver, spleen, 4 large abnormal yolks	Four abnormal yolks	*Bact. sanguinarium*
464	++	∓∓	Active	1 small yolk, capsule not filled, contents thick yellow liquid	Liver, spleen, abnormal yolk, normal yolk, ovary	Abnormal yolk	*Bact. sanguinarium*
465	+∓	+∓	Dormant	Hazelnut-size, greenish-brown yolk; capsule not filled. Several small caseated ovules	Liver, spleen, abnormal yolk, ovules	Abnormal yolk	*Bact. sanguinarium*
477	∓∓	∓−	Dormant	Hazelnut-sized irregular-shaped yolk with opaque capsule, containing yellow pasty material	Liver, spleen, abnormal yolk	Abnormal yolk	*Bact. sanguinarium*
479	++	∓∓	Active	1 large yolk, capsule not filled, contents greyish-yellow thick liquid; 1 large angular yolk, contents partially solidified	Liver, spleen, normal yolks, abnormal yolks	Abnormal yolks	*Bact. sanguinarium*

the losses at that time. However, since *Bact. sanguinarium* is frequently found associated with ruptured yolk, and ovarian infection with the organism was found to exist in the flock, it seems possible that *Bact. sanguinarium* was present at the time the deaths from ruptured yolk occurred and that the ovarian infection may have become established at that time.

Bact. sanguinarium was isolated from abnormal yolks, ovules, or cysts in all except 2 of the 29 birds. Included with those from which *Bact. sanguinarium* was isolated were the three birds (Nos. 349, 350, 423) whose blood serum had given a positive agglutination reaction with *Bact. pullorum* antigen and a negative reaction with *Bact. sanguinarium* antigen. The abnormalities of the ovaries in the two birds (Nos. 304 and 395) from which *Bact. sanguinarium* was not isolated, were very slight, consisting of one small, bloody ovule in bird No. 304 and two small, flabby yolks in bird No. 395. Failure to isolate *Bact. sanguinarium* from these two cases does not necessarily prove, however, that the organism was not present, since it is possible that it was present and that we failed to recover it in cultures.

GENERAL DISCUSSION

These studies demonstrate that *Bact. sanguinarium* may produce an acute, highly fatal disease of young chicks and a chronic infection of the ovaries of hens which cannot be differentiated from disease of chicks and ovarian infection of hens caused by *Bact. pullorum*, except by the difference in the cultural characteristics of the organism isolated from affected birds. Agglutinins occur in the blood serum of hens that are infected with *Bact. sanguinarium*. However, the ordinary routine agglutination test does not serve to differentiate between ovarian infection with *Bact. sanguinarium* and *Bact. pullorum* because serum from a hen infected with the former will cause agglutination of antigens prepared from either of the two species of organisms. This cross-agglutination makes it possible to detect carriers of either *Bact. sanguinarium* or *Bact. pullorum* by an agglutination test employing *Bact. pullorum* antigen. It may, therefore, be considered as enhancing rather than detracting from the value of the agglutination test in the detection of adult hens that harbor infection that may be transmitted through the medium of eggs to offspring.

Although *Bact. sanguinarium* was not isolated from eggs laid by infected hens, these studies furnish evidence that this organism, like *Bact. pullorum*, is transmitted directly to chicks through eggs laid

by infected adults. Deaths among the chicks from *Bact. sanguinarium* infection began when they were 60 hours old. It seems quite certain that the chicks did not acquire the infection from contaminated brooder houses or hovers and very unlikely that the infection originated in the incubators or shipping boxes. The only remaining source of infection is the parent stock among which chronic ovarian infection with *Bact. sanguinarium* was found to exist. It seems probable, therefore, that some of the eggs laid by these hens contained *Bact. sanguinarium,* which resulted in infection of the chicks hatched from them.

The origin of chronic infection of the ovaries of the hens with *Bact. sanguinarium* remains undetermined. It has been found that chicks that survive an outbreak of disease due to infection with *Bact. pullorum* may continue to harbor the infection and that it usually becomes localized in the ovaries. Observations made on 25 of the 66 chicks that survived the infection with *Bact. sanguinarium* and were kept for one year, however, failed to show that any of them had become carriers of the organism. It is in this particular only that these studies fail to show that the behavior of *Bact. sanguinarium* in either chicks or adult fowls may be the same as that of *Bact. pullorum.* The facts that losses from ruptured yolk had occurred in the flock of hens a year before the studies herein reported were made and that *Bact. sanguinarium* infection is frequently associated with ruptured yolk, suggest that the ovarian infection with *Bact sanguinarium* may have then become established. However, no bacteriological examination of dead birds from this flock had ever been made and it was not known that *Bact. sanguinarium* existed in it before the agglutination tests were made.

BIBLIOGRAPHY

[1] KLEIN, E.
1889. Über eine epidemische Krankheit der Huhner verusacht durch einem Bacillus—*Bacillus gallinarum*. Centralbl. f. Bakteriol. 5: 688–693.

[2] MOORE, V. A.
1896. Infectious leukemia of fowls. U. S. D. A., Bur. Animal Ind. Annual Rept. 1895–1896: 187–205.

[3] TAYLOR, W. J.
1916. An outbreak of fowl typhoid. Jour. Amer. Vet. Med. Assoc., 49: 35–49.

[4] SMITH, T., AND C. TENBROECK
1915. A note on the relation between *B. pullorum* (Rettger) and the fowl typhoid bacillus (Moore). Jour. Med. Res. 31: 547–555.

[5] RETTGER, L. F., AND S. A. KOSER
1917. A comparative study of *Bact. pullorum* (Rettger) and *Bact. sanguinarium* (Moore). Jour. Med. Res. 35: 443–458.

[6] GOLDBERG, S. A.
1917. A study of the fermenting properties of *Bact. pullorum* (Rettger) and *Bact. sanguinarium* (Moore). Jour. Amer. Vet. Med. Assoc. 51: 203–210.

[7] HADLEY, P. B.
1918. The colon-typhoid intermediates as causative agents of disease in birds. Rhode Island Agr. Exp. Sta. Bul. 174: 1–216.

[8] PANISSET, M. L., AND J. VERGE
1924. Sur une epizooti de diarrhee blanche bacillaire des poissons. Rev. Gen. Med. Vet. 23: 19–21.

[9] BEAUDETTE, F. R.
1925. The possible transmission of fowl typhoid through the hen's egg. Jour. Amer. Vet. Med. Assoc. 20: 741–745.

[10] DOYLE, T. M.
1926. The method of transmission of avian typhoid. Jour. Comp. Path. and Therap. 39: 137–140.

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POLLINATION AND LIFE HISTORY STUDIES OF LETTUCE (LACTUCA SATIVA L.)

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INTRODUCTION

The lettuce crop is a very important one in California, and the product is shipped in considerable quantity throughout most of the year. The production of lettuce seed is also an important industry. While the variety New York is the only one produced for shipping fresh, almost all of the important varieties are grown for seed. The growing of lettuce for seed is confined almost entirely to the delta lands of the Sacramento and San Joaquin rivers and to the Santa Clara Valley. Wherever a large number of varieties are grown in close proximity, there arises constantly the question of the danger of cross pollination. Pollination investigations reported herein were initiated because of this question. The morphological studies reported in this paper were commenced to furnish information needed in order to prosecute more successfully other lines of investigation that have been started on lettuce.

MATERIAL AND METHODS

The lettuce seed used for growing the material for the morphological studies herein reported, was planted at the University Farm, Davis, California, in December, 1923. The variety employed was Iceberg. The material for study was killed in formalin-alcohol solution, then dehydrated and embedded in paraffin according to the usual procedure. Most of the sections were stained in Delafield's haematoxylin, or safranin and gential violet.

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DEVELOPMENT OF THE FLOWER

In order to allow the flowering stem to develop normally it is often necessary to quarter the heads or else remove the upper leaves entirely. This operation is usually performed before the head becomes hard and while the stems are short (fig. 1). If quartering is delayed until the stem has become elongated (fig. 2) there is danger of injuring it.

The oldest flower head terminates the main axis (fig. 3). The meristematic region of the receptacle, at first convex (fig. 4), becomes flattened and broadened (fig. 5) ; after this there arise, simultaneously over the entire surface, the protuberances that give rise to the individual flowers (figs. 3, 6, 9). The protuberances soon become angular in outline and the appearance of a marginal ring on each protuberance indicates the beginning of the corolla tube (figs. 3, 7, 10). Swellings soon appear upon the inside of the corolla tube (figs. 8, 11) and an elevated ring of meristematic tissue which gives rise to the pappus is formed near the base of the corolla tube and on its outer side (figs. 8, 11). The stamens and pappus appear almost simultaneously. These observations coincide with those of Martin[7] on *Aster* and *Solidago*. The carpels are the last organs of the flower to differentiate (figs. 12, 13). By the time the carpels appear, the corolla has started to curl inward over the top of the flower (fig. 12). The cavity of the ovary is soon formed as a result of the upward growth of the carpellary tissue (fig. 13).

OOGENESIS

The ovule arises at the bottom of the ovarian cavity. The hypodermal archesporial cell, which in this case is the megaspore mother cell, is differentiated early in the life of the ovule (fig. 14). It is easily distinguished by its large size and its deeply staining contents, and by its inclusion within a single-layered nucellar tissue. After the appearance of the megaspore mother cell, the single massive integument becomes increasingly evident (figs. 15, 16). The growth of the integument is unilateral and as a result the ovule finally assumes a position of complete anatropy (fig. 19). The nucellar cells divide anticlinally only, thus allowing for the growth of the megaspore mother cell. The nucellus never consists of more than a single layer of cells. The ovule is completely inverted by the time the heterotypic divisions are initiated.

By two successive divisions (figs. 20, 21, 22, 23) the four megaspores are formed. A wall forms between the two daughter nuclei after the completion of the heterotypic division. Walls are again formed at the completion of the homotypic divisions giving a linear series of four megaspores of equal size. The three micropylar megaspores soon degenerate as a result of encroachment upon them by the functioning innermost megaspore (fig. 26). Virtually the same method of megaspore development has been figured by Merrell[8] for *Silphium,* by Small[13] for *Senecio,* and by Martin[7] for *Aster* and *Solidago.* The nucellar cells abutting on the micropyle appear to enlarge slightly for a time (fig. 26). The rapidly growing embryo sac, however, soon crowds the lower cells of the nucellus and the degenerating megaspores into the upper end of the micropyle to form the so-called nucellar cap (fig. 45).

The functioning or fertile megaspore enlarges considerably before the first division of its nucleus (fig. 26). The first nuclear division takes place near the middle of the embryo sac (fig. 27). The daughter nuclei then migrate toward opposite ends (figs. 28, 29) and by two successive divisions form four free nuclei at the extremities of the sac. The polar nuclei move toward the center and finally come to rest just above the egg (fig. 32).

The polar nuclei are rather distant from one another two hours before anthesis (fig. 32) and, so far as has been determined, fusion of the polar nuclei does not take place until the time of fertilization. According to Land,[6] in *Silphium* the polar nuclei fuse long before fertilization, whereas Nawaschin[10] reports their early fusion in *Helianthus.*

Not more than three antipodal cells were ever observed. Sometimes they are in a linear row completely filling the antipodal neck of the embryo sac (fig. 62) while in other cases, the two lower antipodals lie side by side with the third cell forming a sort of summit. The variability in number reported for *Silphium,*[8] *Aster* and *Solidago,*[2] *Senecio,*[9] *Erigeron,*[6] and *Bellis*[1] does not seem to occur in *Lactuca.* Schwere[12] reports only three antipodals for *Taraxacum* and, according to Small,[13] this number seems to be the rule for most of the *Cichoriaceae.*

The occurrence of two ovules in a single ovary is common (fig. 25), and often three ovules were observed. Occasionally an ovule was observed with two developing embryo sacs (fig. 25), but in no case were two embryos observed within the same ovule.

A single layer of inner integumentary cells forms a jacket completely surrounding the embryo sac. This layer is very conspicuous

before the egg apparatus is mature (fig. 25) and does not entirely disappear until the seed is almost ripe. According to Coulter and Chamberlain,[3] this layer of cells is nutritive in function. These authors also mention a large number of genera in which this nutritive jacket has been observed.

SPERMATOGENESIS

When the young stamens have reached the stage of development shown in figures 34 and 35, a single row of hypodermal archesporial cells is already distinguishable. The first periclinal wall cuts off a single row of pollen mother cells on the inside (fig. 36), and an outer layer, which divides to form the tapetum, the middle layer, and the endothecium (fig. 37). The pollen mother cells and their nuclei enlarge considerably before the heterotypic division is initiated (fig. 37).

Gates and Rees[4] made a cytological study of pollen development in three species of *Lactuca: L. sativa* L., *L. scariola* L., and *L. muralis* Frees. In a single loculus of each species, they found from fifteen to twenty pollen mother cells, which in turn produce a total of about sixty pollen grains. According to these authors, the pollen mother cells frequently separate from each other before synapsis. Figure 38 shows the pollen mother cells separated and well rounded off. About the beginning of synizesis, or a little earlier, the tapetal cells become binucleate. This latter stage is well illustrated in figure 37. Later the tapetal cells become quadrinucleate (fig. 39).

According to Gates and Rees[4] there are nine bivalent chromosomes. They state that at the time of diakinesis "the nine bivalent chromosomes form a graded series which can be arranged in a general way in three groups, three of maximum length four or five times as long as broad, three of intermediate length, about two or three times as long as broad, and three very short and almost cubical. Between the stage of diakinesis and the arrangement of the chromosomes on the heterotypic metaphase they are condensed to such a degree that there remains little observable difference in length between them."

In the homotypic metaphase (fig. 41) the spindles extend in opposite planes. According to Coulter and Chamberlain,[3] this method of division is common in the dicotyledons; the two nuclear divisions occur before the walls are formed, then all of the latter are formed simultaneously. The method of cytokinesis in *Lactuca* has been studied in detail by Gates and Rees,[4] who state that "after the

reduction divisions the cytoplasm of the pollen mother cells begins to constrict at four points and these constrictions finally meet at the center, cutting the contents of the cell into four parts. The young pollen grains so formed alter their shape within the mother-cell wall, becoming roughly heptagonal and then secreting a cell wall. The mother-cell wall then breaks down and the wall of the pollen grain ultimately becomes remarkably thickened and sculptured."

The pollen grain germinates within the anther. Samples collected several hours before anthesis (fig. 44) show two filamentous sperms and a vegetative nucleus. The sperms at this time reach about half way around the interior wall of the pollen grain. Filamentous sperms of this general type in the Compositae have been described by Merrell[8] and Land[6] for *Silphium*, and by Nawaschin[10] for *Helianthus*.

Figure 24 shows a transverse section of a flower head, the anthers of which are in the mother-cell stage. This figure shows a large number of individual flowers that have more or fewer stamens than the characteristic number, five. The syngenesious character of the androecium is not well illustrated in this figure.

POLLINATION

About twenty-four hours before anthesis, the bracts subtending the flower head start to open at the summit because of the development of the individual flower buds (fig. 84). The buds make a remarkable elongation during the twenty-four hours before anthesis. The extent of this development can best be seen by comparing figure 84 with figure 87. Figure 84 is taken from the head illustrated in figure 83. Figure 87 shows the flower at time when the floral organs are fully extended and expanded and the brush hairs of the pistil are covered with pollen. Figure 85 shows the flower head two hours before anthesis. In figure 86 the pistil is not fully extended; the brush hairs that are visible are covered with pollen; and the stigmatic lobes have started to expand.

Although the lettuce flower is almost entirely self-pollinated, crossing can take place. When the pistil elongates and pushes its way through the anther tube, the brush hairs on the side of the pistil sweep the pollen grains upward out of the pollen sacs of the dehisced anthers. According to Oliver,[11] the anthers dehise before the flower head opens and when the unexpanded stigmas appear through the staminal column they are already covered with pollen. When the stigmatic

lobes expand, pollen falls on the stigmatic hairs of the inner surface, insuring self-pollination. Knuth[5] states that when the stigmatic lobes expand, they make a complete revolution backwards and the stigmatic papillae come into contact with the pollen held by the brush hairs of the pistil, thus bringing about self-pollination. It is not known definitely whether or not the pollen grains will germinate elsewhere on the pistil than on the stigmatic papillae.

Oliver[11] suggests that crossing may occur in the field if the pollen has been washed from the stigmas by rain. Foreign pollen may then be brought in by insects. Since rains occur so seldom in California during the lettuce blooming season, the danger from this source is almost negligible. If self-pollination is brought about, in the manner described by Knuth,[5] there is danger of foreign pollen being brought in by insects before the inner surface of the outward curving stigmatic lobes have come into contact with the pollen held by the brush hairs of the pistil of the same flower. There is also the possibility of differential pollen-tube growth between foreign pollen and that of self-pollen.

Observations were made on methods of natural pollination in the White Paris Company's variety of lettuce in June, 1926, at Davis, California. A number of flower heads were protected against insect visitation. When the stigmatic lobes were fully expanded the pollen grains that had fallen on the inner surface of the stigmatic lobes of each flower were counted. Seventy flowers were observed. Of these 58 did not have any pollen grains on the inner stigmatic surfaces. The number of pollen grains on the inner stigmatic surfaces of the other 12 flowers ranged from 1 to 7. The edges of the stigmatic lobes were always covered with pollen, however, as well as the backs of the lobes. It was also observed that only occasionally do the stigmatic lobes make a complete revolution backwards.

Even though the anthers dehisce and the pollen grains are in contact with the stigmatic lobes and upper portion of the style before the latter is extended through the staminal column, Oliver[11] states he was able to completely depollinate the lettuce flowers by washing off the pollen with a fine stream of water from the fully extended pistils. This method of depollination has been used by a number of investigators.

With reference to pollination by insects, Knuth[5] reports different species of flies visiting the flowers. Flies have also been observed visiting the flowers in various lettuce seed fields in California. At Davis, California, a brilliant green bee, *Agapostemon texanus* Cresson[2] was

[2] Identified by Professor T. D. A. Cockerell.

found to be the most frequent insect visitor. Several species of *Halictus*[3] have also been observed visiting the flowers. The bees that visit the lettuce flowers are probably pollen collectors, only. In June, 1926, a count was made of the number of pollen grains on the surface of the stigmatic lobes of flowers that were insect visited. All of the seventy flowers observed had pollen grains on the inner stigmatic lobes. These ranged in number from 4 to 51. One plant observed had 169 flower heads open on June 26. During the short time that these were open 88 were visited at least once by insects and some were visited a number of times. On the same day another plant had 82 flower heads open and 60 of these were visited by insects.

As a protection against insect visitation and to avoid the danger of introduction of foreign pollen, some seedsmen enclose the selected plants in cloth bags during the entire flowering period. These bags may be left on the plants until the seed is harvested.

The stigmatic papillae develop from large rectangular cells that are very early differentiated. At an early stage of development, these cells stain very deeply and possess exceptionally large nuclei. In figure 33, these large cells are seen abutting on the nutritive cells below. These nutritive layers coalesce below the style branches and form a single tissue through the style. A single vascular bundle is found in each lobe of the stigma. At the stage shown in figure 33, the microspores have already escaped from the wall of the pollen mother cell and the outer sculpturing is beginning to form. Figure 31 shows the stigmatic papillae somewhat elongated. At this stage the embryo sac is two-nucleate, as shown in figure 29.

ANTHESIS

In 1925, studies were made of a number of lettuce plants in order to determine the waves of anthesis occurring during the season and the length of time required for the seeds to ripen. Seed of the Iceberg variety was planted in November, 1924. In the early spring of 1925, when the plants were about two inches high, they were thinned so that they were twelve inches apart in the row. The heads were quartered while still soft so as to allow the emergence of the seed stem. Ten plants that had flowers in bloom for the first time on June 11 were selected for study. Table 1 gives the number of flowers in bloom each day on the different plants from June 11 to July 30. The taking of

[3] Miss Grace Sandhouse of the U. S. National Museum identified one species of *Halictus* as *H. titusi* Crawford.

records was discontinued after July 30. The flower heads were counted each morning at the time of anthesis.

Lettuce plants show definite flowering peaks. The data in table 1 show that almost all the plants reached a flowering peak in the latter part of June. Then a drop to zero occurred in some cases, continuing for several days, and then another peak of less magnitude occurred in July. The general flowering curve and its minor irregularities due mainly to fluctuations in temperature can best be studied when individual plants are considered. The morning after a very warm day there is usually a decided rise in the flowering curve; the morning after a cool day, there is usually a pronounced drop. The influence of temperature is more noticeable during the early than the latter part of the flowering period. Although different plants may start flowering at the same time, their flowering curves seldom parallel one another. Some plants also have more definite flowering cycles than others. Those of plants 5 and 7, are not nearly so pronounced as those of plants 1, 2, 3, 6, 8, and 9. In figure 109 the flowering and seed ripening curves are plotted for plant No. 8. The mean temperature for the same period is also given.

RIPENING OF THE ACHENES

The lettuce flowers are open usually an hour or two only. The ligulate corollas then fold tightly together and do not again open (fig. 88, 89). Within two or three days the corollas, dehisced stamens, and withered styles and stigmas are shed in a cluster. The bracts then close tightly about the developing achenes. This is well illustrated in samples collected three days after anthesis (fig. 91). The beak of the young fruit elongates and carries the pappus along in its upward growth. The rate of elongation is fairly rapid, as can be observed in the different aged fruits in figures 89, 90, 92, 93, and 95. These illustrations were made from samples collected 1, 2, 3, 4, and 5 days after anthesis. Within four or five days after anthesis, the pappus begins to appear through the bracts (fig. 94). The growth made by the embryo from ten hours after anthesis to the morning of the eleventh day are shown in figures 96 to 108C. The embryo in each figure has been colored black. The intervals of time between anthesis and collection of the samples used for the illustrations are as follows: figure 96, ten hours; figure 97, fourteen hours; figure 98, twenty-six hours; figure 99, thirty-four hours; figure 100, three days; figure 101, four days; figure 102, five days; figure 103, six days;

TABLE 1

LETTUCE ANTHESIS. 1925

Date	Number of flower heads in bloom										
	Plant No. 1	Plant No. 2	Plant No. 3	Plant No. 4	Plant No. 5	Plant No. 6	Plant No. 7	Plant No. 8	Plant No. 9	Total	Average
June 11	1	1	1	1	1	4	3	2	2	16	1.8
12	9	1	2	3	0	9	5	4	0	33	3.7
13	20	9	8	11	0	19	11	4	3	85	9.4
14	51	8	27	19	3	45	21	13	2	189	21.0
15	59	24	31	68	2	33	30	18	3	268	29.8
16	33	21	27	43	4	16	28	26	4	202	22.4
17	39	33	21	34	10	33	20	64	10	264	29.3
18	48	33	25	24	7	61	23	63	12	296	32.9
19	71	25	41	64	5	86	20	65	24	401	44.6
20	69	21	85	108	16	82	23	95	43	542	60.2
21	55	27	117	101	61	72	21	117	67	638	70.8
22	36	48	91	79	46	49	19	109	48	525	58.4
23	30	43	45	24	33	49	22	85	30	361	40.1
24	27	57	69	42	67	51	34	116	52	515	57.2
25	22	51	53	27	82	53	21	145	107	561	62.4
26	22	85	65	50	109	68	43	196	154	792	88.0
27	61	100	84	47	96	72	52	170	140	822	91.4
28	40	75	91	41	70	62	31	133	74	617	68.5
29	24	77	69	37	47	34	20	80	65	453	50.3
30	14	42	46	19	28	10	26	39	38	262	29.1
July 1	9	28	50	32	38	6	25	14	16	218	24.2
2	5	22	42	23	48	1	32	2	15	190	21.1
3	6	10	9	17	32	1	22	0	6	103	11.4
4	1	4	4	14	34	0	25	0	6	88	9.8
5	0	8	8	16	43	0	22	0	12	109	12.1
6	3	7	10	9	30	0	19	2	8	88	9.8
7	2	7	11	18	36	0	25	1	17	117	13.0
8	3	7	13	24	40	0	19	0	24	130	14.4
9	3	17	6	34	26	2	29	2	23	142	15.8
10	4	25	25	29	31	5	29	11	31	190	21.2
11	5	40	23	49	37	5	25	25	34	243	27.0
12	3	37	31	43	30	13	35	24	31	247	27.5
13	21	51	26	67	27	19	39	28	16	294	32.7
14	27	45	52	72	23	25	49	44	16	353	39.2
15	38	43	58	64	28	38	50	93	41	453	50.3
16	61	58	72	46	46	35	52	121	60	551	61.2
17	55	46	63	25	43	46	53	103	41	475	52.8
18	20	41	50	9	36	47	31	67	38	339	37.6
19	7	57	40	.....	27	54	25	83	62	355	44.4
20	2	44	31	.....	18	52	9	71	35	262	32.7
21	3	49	26	.....	22	35	12	64	41	252	31.5
22	2	28	16	.....	7	26	4	32	21	136	17.0
23	0	31	15	.....	17	16	2	39	33	153	19.1
24	1	46	20	.....	4	11	0	35	16	133	16.6
25	1	37	20	.....	12	20	1	42	12	145	18.1
26	4	43	14	.....	8	12	0	19	18	118	14.8
27	2	37	14	.....	15	8	0	27	21	124	15.5
28	3	26	7	.....	12	10	0	27	11	96	12.0
29	9	35	21	.....	11	21	0	40	19	156	19.5
30	13	50	19	.....	17	37	0	55	27	218	27.0
Total.....	1,044	1,760	1,794	1,433	1,485	1,453	1,107	2,615	1,629	14,320	

figure 104, seven days; figure 105, eight days; figures 106 and 108A, B, and C, eleven days; and figures 107A, B, and C, nine days. On the twelfth day (June 24, 1924) the seeds were ripe. The long filamentous suspensor is very conspicuous in samples collected three and four days after anthesis (figs. 100, 101).

In 1925 seed-ripening studies were made on the same plants that were used for a study of flowering habit.

TABLE 2

SEED-RIPENING DATA. 1925

Date	Seed heads mature on plant No.										
	1	2	3	4	5	6	7	8	9	Total	Average
June 22	0	0	0	0	0	4	0	0	0	4	.4
23	2	3	0	0	1	0	3	0	2	11	1.2
24	11	0	2	2	0	9	5	4	0	33	3.7
25	48	12	12	8	0	33	16	7	3	139	15.4
26	86	24	41	41	4	60	55	32	6	349	38.8
27	37	20	30	49	2	37	25	39	5	244	27.0
28	42	41	33	64	12	6	20	55	13	286	31.8
29	43	21	22	30	3	84	22	48	12	285	31.8
30	13	13	33	32	6	27	6	15	10	155	17.2
July 1	60	21	32	38	4	81	15	55	18	324	36.0
2	66	23	58	33	19	96	21	100	47	463	51.4
3	62	44	60	144	84	91	35	194	89	803	89.2
4	25	24	60	66	19	17	4	23	19	257	28.5
5	29	49	51	45	60	56	34	94	54	472	52.4
6	15	36	37	11	37	28	18	69	35	286	31.8
7	10	37	27	15	72	33	21	62	60	337	37.4
8	20	62	57	41	59	62	24	93	91	509	56.5
9	28	76	69	40	95	65	42	172	130	717	79.8
10	38	64	69	38	67	49	28	195	93	641	71.2
11	47	76	100	40	67	65	46	136	87	664	73.8
12	.....	.....	.....	50	57	39	49	119	92	406	67.6
13	.....	.....	.....	9	17	0	7	7	17	57	9.5
14	.....	.....	.....	18	43	5	30	11	18	125	20.8
15	.....	.....	.....	19	44	1	19	5	14	102	17.0
16	.....	.....	.....	20	40	0	15	4	8	87	14 5
17	.....	.....	.....	27	60	0	50	9	23	169	28 1
18	.....	.....	.....	.....	44	0	27	1	12	84	16.8
19	.....	.....	.....	.....	68	0	37	6	49	160	32 0
20	.....	.....	.....	.....	41	8	28	16	36	129	25.8
21	.....	.....	.....	.....	36	7	45	21	36	145	29.0
22	.....	.....	.....	.....	19	6	17	18	25	85	17.0
23	.....	.....	.....	.....	24	8	29	21	19	101	20 5
24	.....	.....	.....	.....	20	9	35	22	18	104	20.8
25	.....	.....	.....	.....	11	15	22	34	15	97	19.4
26	.....	.....	.....	.....	16	5	44	20	8	93	18.6
27	.....	.....	.....	.....	16	22	12	44	29	123	24.6
28	.....	.....	.....	.....	31	32	43	98	45	249	49.8
29	.....	.....	.....	.....	32	36	47	67	36	218	43.8
Total	682	646	793	880	1,230	1,096	996	1,916	1,274	9,513	

Each day all the seed heads which had ripened their achenes were counted and then removed. The first seed heads were ripe on June 22, as is shown in table 2. Of the nine plants under observation only one ripened seed on this day. Some plants did not ripen seed until June 24. The average number of flower heads open and the average number of seed heads ripe on the different dates from June 11 to July 30 are plotted in figure 110. It is thus seen that the two high peaks in seed ripening occurred twelve days after the flowering peaks. This indicates that the average time from anthesis to fruit maturity is about twelve days, under conditions as they existed at the time. If the temperature remains high, the ripening of the achenes is hastened, and if the average temperature is low, the time from anthesis to fruit maturity is increased.

In counts made on thirty different heads, which were taken from several plants, it was found that the number of flowers varied from 15 to 22, with an average of 18.3. The number of normally developed achenes averaged 16.2 to a head. Either a number of the ovules are not fertilized or the embryos fail to develop.

FERTILIZATION

In June, 1924, a study was made to determine the length of time between pollination and fertilization and also the interval between pollination and fruit maturity, and other attendant phenomena. The study was started the morning of June 12. Table 3 gives a record of temperatures taken in the shade of lettuce plants in the field.

TABLE 3

TEMPERATURES IN THE SHADE. JUNE 12, 1924

Time	Temperature degrees C.	Time	Temperature degrees C.
5 A.M.	12.0	1 P.M.	31.0
6 A.M.	13.5	2 P.M.	31.8
7 A.M.	16.0	3 P.M.	31.4
8 A.M.	21.5	4 P.M.	28.8
9 A.M.	24.0	5 P.M.	27.0
10 A.M.	26.5	6 P.M.	25.0
11 A.M.	28.6	8 P.M.	18.0
Noon	29.4	10 P.M.	14.5

At 5 A.M. and at 6 A.M. the flowers were still closed. At 7 A.M. the corollas were just starting to unfold, and although the stigmas were still enclosed within the staminal column, some of the anthers had

dehisced. The flowers were fully expanded at 8 A.M. and the pistils were fully extended. Eight o'clock is hereafter referred to as the time of pollination in this study.

At 11 o'clock the same morning sperms were first observed in the embryo sac. There were only a very few embryo sacs that contained sperms at this time. At noon a few more of the embryo sacs had sperms in them (fig. 46). By 1 P.M., or five hours after pollination, the majority of the embryo sacs contained sperms. In samples taken at 2 P.M., no fertilization stages were found, but the embryo sacs contained all stages of development from fertilized eggs to two-celled embryos (figs. 47 to 50). The time elapsing between pollination and fertilization in lettuce is, then, extremely short. Considering 8 A.M. as the time of pollination, it was only three hours before the first fertilization stages were observed, and in less than six hours, fertilization had been completed in all the flowers studied. In figure 45, one male cell is lying near the egg nucleus. The polar nuclei are fusing, but the second sperm destined to unite with the two polar nuclei can not be detected. In figure 46, one male cell is in close proximity to the egg nucleus, and the other is in contact with the polar nuclei. It appears that there is approximately simultaneous union of the three nuclei to form the primary endosperm nucleus.

DEVELOPMENT OF THE EMBRYO

The embryo of lettuce follows the general type of development outlined by Carano[1] and Souèges[14] for other members of the Compositae. The term embryo instead of proembryo is used in this discussion.

After fertilization, the zygote develops a definite wall and elongates considerably. The lower or micropylar end is occupied by a large vacuole. The large nucleus usually contains two or three nucleoli (fig. 47). Divisions are initiated very shortly after fertilization. A number of two-celled embryos were found at 2 P.M., three hours after the first sperms were observed in the embryo sacs. The first wall is transverse. This wall cuts off a terminal cell (fig. 50, *a*), from which develop the cotyledons and epicotyl, as well as a lower cell (fig. 50, *b*), which gives rise to the hypocotyledonary parts of the embryo. The upper cell (*a*) is much the smaller. The basal cell (*b*) has a large vacuole occupying the greater portion of the lower two-thirds, thus forcing the nucleus to occupy a terminal position. Both cells of the two-celled embryo divide at very nearly the same time. In figure 51,

both cells are in anaphase, but cell *a* is slightly in advance of cell *b*. In figure 52, cell *a* is in telophase while cell *b* is still in metaphase. While the basal cells of these two figures are in the same stage of development, cell *a* of figure 53 is somewhat in advance of that of figure 52. In figure 54, cell *a* is in telophase and cell *b* is in anaphase. The upper cells of figures 53 and 54 are in approximately the same stage of development, but the basal cell of figure 54 is slightly farther advanced than that of figure 53. In figure 55, there are two cells in tier *a*; cell *b* is still in telophase. Thus, it is seen that the division of cell *b* lags slightly behind that of cell *a*, and as a general rule, the cells at the terminal portion of the embryo develop more rapidly than those of the basal part. In the four-celled embryo, the two cells in tier *a* are formed by a longitudinal wall, while cells *c* and *d*, daughter cells of *b*, are formed by a transverse wall (fig. 56, 57). The embryo shown in figure 56, is as it appears only 6 or 7 hours after fertilization.

Every cell of the four-celled embryo contributes to the formation of the eight-celled embryo. Again, each tier usually divides slightly in advance of the one below it. In figure 58, both cells of tier *a*, and also of cell *c*, are in metaphase, while cell *d* is still in prophase. In figure 59, both cells of tier *a* are in anaphase, one slightly in advance of the other; cell *c* and cell *d* are in prophase. In figure 60, cell *c*, which is in anaphase, is slightly in advance of both cells of tier *a*, which are in metaphase, and of cell *d*, which is in prophase. In figure 61, one cell of tier *a* is in anaphase while the other is in metaphase; cell *c* is in mid-anaphase while cell *d* is in prophase. The eight-celled embryo of figure 63 was observed in samples collected 20 hours after pollination. In this figure, tier *a* has four cells, and tier *c*, two cells; *e* and *f* are daughter cells of *d*. The quadrants of tier *a* are formed by the development of two longitudinal walls at right angles to the initial wall. A longitudinal wall divides tier *c*, but *e* and *f* have been formed from *d* by a transverse wall. Thus the three lower tiers of cells in the eight-celled embryo (fig. 63) have been derived from the basal cell, and the upper tier of four cells has been derived from the terminal cell of the two-celled embryo. While the general plan of development of the young embryo is well established, the relative rates of development of similar cells in the different embryos are not always the same. Doubtless the rates of development and of division of the different cells are considerably influenced by the conditions that surround them.

The sixteen-celled embryo is formed by the division of each cell of the eight-celled embryo. The cells of tier *a* are the first to divide. The wall dividing each of the quadrant cells of tier *a* is united with

the peripheral wall near its middle (fig. 64), and with the lower horizontal wall. Occasionally this wall is attached to the base of the vertical walls (fig. 71). When the octants are formed in tier *a*, the embryo is twelve-celled (fig. 64). No more divisions occur in this layer until all the tiers of cells below it have undergone division. Tier *c* is the next to divide. In figure 64, both cells of tier *c* are in anaphase and cell *e* in prophase. Segmenting walls develop at right angles to the axial wall and divide tier *c* into quadrants forming the fourteen-celled embryo (fig. 65). Cell *e* divides later by a vertical wall forming the fifteen-celled embryo (figs. 66–71). The division of cell *f* lags considerably behind that of cell *e*. In figure 68, cell *f* is in prophase, in figures 69 and 70, it is in metaphase, and in figure 71, it is in telophase. The division of cell *f* by a transverse wall completes the sixteen-celled embryo (fig. 72). In the sixteen-celled embryo, the eight cells of the upper tier are derived from the terminal cell of the two-celled embryo, while the total of eight cells in tiers *c*, *e*, *g*, and *h* have been derived from the basal cell. In the sixteen-celled embryo, then, as many cells have been derived from the basal as from the terminal cell of the two-celled embryo. This stage was found in samples collected 25 hours after pollination. The early development of the embryo is very similar to that described by Soueges for *Senecio*. In figure 73, tangential walls have differentiated the dermatogen, periblem, and plerome. In tier *e*, four cells are formed by the development of vertical walls. Tangential walls then differentiate the dermatogen from four inner cells which are the periblem initials. Transverse walls divide the plerome cells of tier *c* into a two-celled layer (figs. 75, 76). After this a series of longitudinal and transverse cell divisions differentiates the plerome of tier *c*, a four-celled layer (fig. 78). This stage of development is reached three days after anthesis and at the same time that the cotyledonary elevations are first noticeable in tier *a* (fig. 78).

It is rather difficult to determine the sequence of wall formation in tier *g*. In figure 76, the first wall appears to be longitudinal, while in figure 77, it appears to be transverse. In samples collected exactly four days after pollination (fig. 79), transverse and longitudinal wall formation had made of tier *g* a two-celled layer, the upper cells of which are the dermatogen initials, and the lower of which contribute to the formation of the root cap. Thus tier *e* contributes the periblem initials and dermatogen cells to the young embryo, and tier *g* contributes the dermatogen initials and a portion of the root cap.

The root cap is built up by the division of the lower dermatogen cells of tier *e*, and from the dermatogen cells of tiers *e* and *g*. The

cells of the root cap are formed by periclinal division of the cells of the dermatogen. The method of formation and the development of the root cap correspond very closely with that described by Carano[1] for *Bellis*.

Tier *c* contributes the plerome initials, one of which is seen dividing in figure 79A. A definite single cell layer, the pericycle, is already differentiated in figure 79A and from all appearances has its own initials.

The dermatogen, periblem, and plerome tissues of the hypocotyl all arise from different initials. As previously stated, the root cap is built up by periclinal division of the lower dermatogen cells and dermatogen initials. The suspensor is formed from cell *h*, and varies in length and in the number and method of division of its cells.

ENDOSPERM

The divisions of the endosperm nuclei are at first slightly in advance of those of the embryo. When the embryo is four-celled, the free endosperm nuclei have almost completed their fourth division (fig. 80). During the early stages, the divisions of the free nuclei are usually nearly simultaneous. This is shown very well in figure 80, where six of the dividing nuclei are in telophase, while the other two are in late anaphase. This parallelism of division, however, is not always maintained, for in figure 57, a transverse section of an embryo-sac containing an embryo of the same age and stage of development as that in figure 80, one endosperm nucleus is in late anaphase while the other appears to be in a resting condition. By the time the eight-celled embryo is formed, twenty hours after pollination, walls have appeared in the endosperm (fig. 81). The endosperm cells at this time completely fill the embryo-sac; they are few in number, very large and highly vacuolate. The endosperm cells continue to divide. They grow with such rapidity for a considerable time as to continue to occupy the space within the rapidly enlarging embryo sac. Figures 98 to 103, showing endosperm development, were drawn from samples collected 26 and 34 hours, and 3, 4, 5, and 6 days after pollination.

The developing embryo rapidly encroaches upon the endosperm, digesting and absorbing it. The seventh day after pollination (fig. 104), the endosperm tissue is almost entirely destroyed. Figure 82, drawn from a sample collected three days after pollination, shows the outer layers of endosperm cells compressed and flattened against the integument. The walls of the two outer cell layers of the endosperm

become somewhat thickened and are very conspicuous during the late stages of seed development (figs. 104 108C). This membrane is also present in the ripe seed and completely invests the embryo.

The endosperm cells do not contain much reserve food at any stage of their development. They are always highly vacuolate. When the seed is ripe, all of the reserve food is stored within the cells of the embryo, and it is upon this food supply that the growing seedling must draw until it starts to manufacture its own food. The nutritive layer of the integument, which is very conspicuous during the early stages of development of the embryo (figs. 81, 82) is gradually absorbed and disappears entirely by the time the seed is mature.

SUMMARY

(1) In the development of the flower, the first primordial whorl to appear is that of the corolla. The primordia of the stamens and pappus are the next to appear, at approximately the same time, while the primordial tissue of the carpels is the last to develop.

(2) The archesporium is the megaspore mother cell. Four megaspores are formed. The inner enlarges to form the embryo-sac. The nucellus consists of a single layer of cells. The polar nuclei appear to unite at about the time of fertilization. Not more than three antipodal cells were ever observed. A nutritive jacket of integumentary cells completely surrounds the embryo-sac.

(3) The development of the microspore precedes that of the megaspore. Very early there is formed a single row of pollen mother cells, each giving rise to four microspores. The tapetum, middle layer, and endothecium arise from the sister cell of the pollen mother cell. At the time of pollination, in addition to the tube nucleus, two sperms, filamentous in form, reach about half way around the interior wall of the pollen grain.

(4) The lettuce flower is almost entirely self-pollinated. Pollen is shed before the flower head is fully expanded. The pollen is carried from the anther tube by the brush hairs on the outside of the elongating pistil.

(5) Lettuce flowers are open for only a short time. The lettuce plant as a whole usually shows definite flowering peaks. Plants under observation at Davis, California, reached a flowering peak in the latter part of June and another of less magnitude in July. Minor irregularities in the flowering curve are due primarily to fluctuations in the temperature.

(6) Under the conditions of this study, the average time from anthesis to maturity of the achenes was about twelve days.

(7) Studies made on June 12, 1924, show that fertilization is complete in less than six hours after pollination. A few sperms were observed in the embryo sacs three hours after pollination. Five hours after pollination sperms were found in most of the embryo sacs studied.

(8) The zygote divides very soon after fertilization. The first wall is transverse, cutting off a small terminal cell which gives rise to the cotyledonary and plumule parts of the embryo and a large basal vesicular cell, from which develops the hypocotyledonary portions. The upper cell divides by a longitudinal wall and the lower by a transverse wall, forming the four-celled embryo. Every cell of the four-celled embryo, by division, contributes to the formation of the eight-celled embryo. In the sixteen-celled embryo, as many cells have been derived from the basal as from the terminal cell of the two-celled embryo. At this stage there are five tiers of cells. The upper tier of eight cells contributes the cotyledons and plumule. The second tier of four cells gives rise to all the primary tissues of the hypocotyl and plerome initials, the third tier of two cells produces dermatogen cells and periblem initials. The fourth tier (one celled) contributes the dermatogen initials and a portion of the root cap, while the fifth tier or basal cell develops into a many-celled filamentous suspensor. Cells of the root cap are formed by periclinal division of the cells of the dermatogen.

(9) The divisions of the free endosperm nuclei at first precede those of the embryo. By the time the embryo is eight-celled, walls have appeared in the endosperm. A two-celled endosperm layer persists in the lettuce seed; all other endosperm tissue is digested and absorbed by the embryo before the latter matures.

LITERATURE CITED

[1] Carano, E.
1914. Richerche sull' embriogenesie delle Asteracee. Ann. di. Bot. 13: 251–301.

[2] Chamberlain, Chas. J.
1895. The embryo-sac of Aster novae-angliae. Bot. Gaz. 20:205–212.

[3] Coulter, J. M., and C. J. Chamberlain
1909. Morphology of angiosperms.

[4] Gates, R. Ruggles, and E. M. Rees
1921. A cytological study of pollen development in Lactuca. Ann. Bot. 35:365–398.

[5] Knuth, Paul
1898. Handbuch der Blütenbiologie. 1–697.

[6] Land, W. J. G.
1900. Double fertilization in Compositae. Bot. Gaz. 30:252–260.

[7] Martin, G. W.
1892. Development of the flower and embryo sac in Aster and Solidago. Bot. Gaz. 17:353–358, 406–411.

[8] Merrell, W. D.
1900. A contribution to the life history of Silphium. Bot. Gaz. 29: 99–133.

[9] Mottier, David M.
1893. On the embryo sac and embryo of Senecio aureus L. Bot. Gaz. 18: 245–253.

[10] Nawaschin, S.
1900. Über die Befruchtungsvorgänge bei einigen Dicotyledonen. Ber. Deutsch. Bot. Gesell. 18:224–230.

[11] Oliver, Geo. W.
1910. New methods of plant breeding. U. S. D. A. Bur. Pl. Ind. Bul. 167:1–39.

[12] Schwere, Siegfried
1896. Zur Entwickelungsgeschichte der Frucht von Taraxacum officinale Web. Ein Beitrag zur Embryologie der Compositeen. Flora 20:32–66.

[13] Small, James
1918–1919. The origin and development of the Compositae. New Phytol. 17:13–40, 69–94, 114–142, 200–230; 18:1–35, 65–90, 128–176.

[14] Souèges, M. Rene
1920. Embryogenie des Composées. Les premiers stades du développement de l'embryon chez le Senecio vulgaris L. Compt. Rend Acad. Sci. Paris 171:254–256.

EXPLANATION OF PLATES

ABBREVIATIONS

m.a., main axis; *t.fl.*, terminal flower head; *r.*, receptacle; *cor.*, corolla; *st.*, stamens; *pa.*, pappus; *carp.*, carpels; *in.*, integument; *nuc.*, nucleus; *m.m.*, megaspore mother cell; *o.c.*, ovarian cavity; *peri.*, pericarp; *m.*, megaspores; *br.*, bracts; *ov.*, ovule; *e.s.*, embryo sac; *micro.*, micropyle; *s.*, style; *anth.*, anther; *p.n.*, polar nuclei; *syn.*, synergids; *n.c.*, nucellar cap; *sp.*, sperms; *v.n.*, vegetative nucleus; *n.j.*, nutritive jacket; *vac.*, vacuole; *t.c.*, tapetal cells; *p.m.c.*, pollen mother cells; *a.c.*, archesporial cells; *epi.*, epidermis; *s.p.*, stigmatic papillae; *te.*, tetrads; *e.n.*, egg nucleus; *der.*, dermatogen; *n.t.*, nutritive tissue; *endo.*, endothecium; *m.l.*, middle layer; *der. i.*, dermatogen initials; *peri.*, periblem; *peri. i.*, periblem initials; *pl.*, plerome; *pl. i.*, plerome initials; *r.c.*, root cap; *peric.*, pericycle; *endo.*, endodermis; *en.*, endosperm; *emb.*, embryo; *susp.*, suspensor.

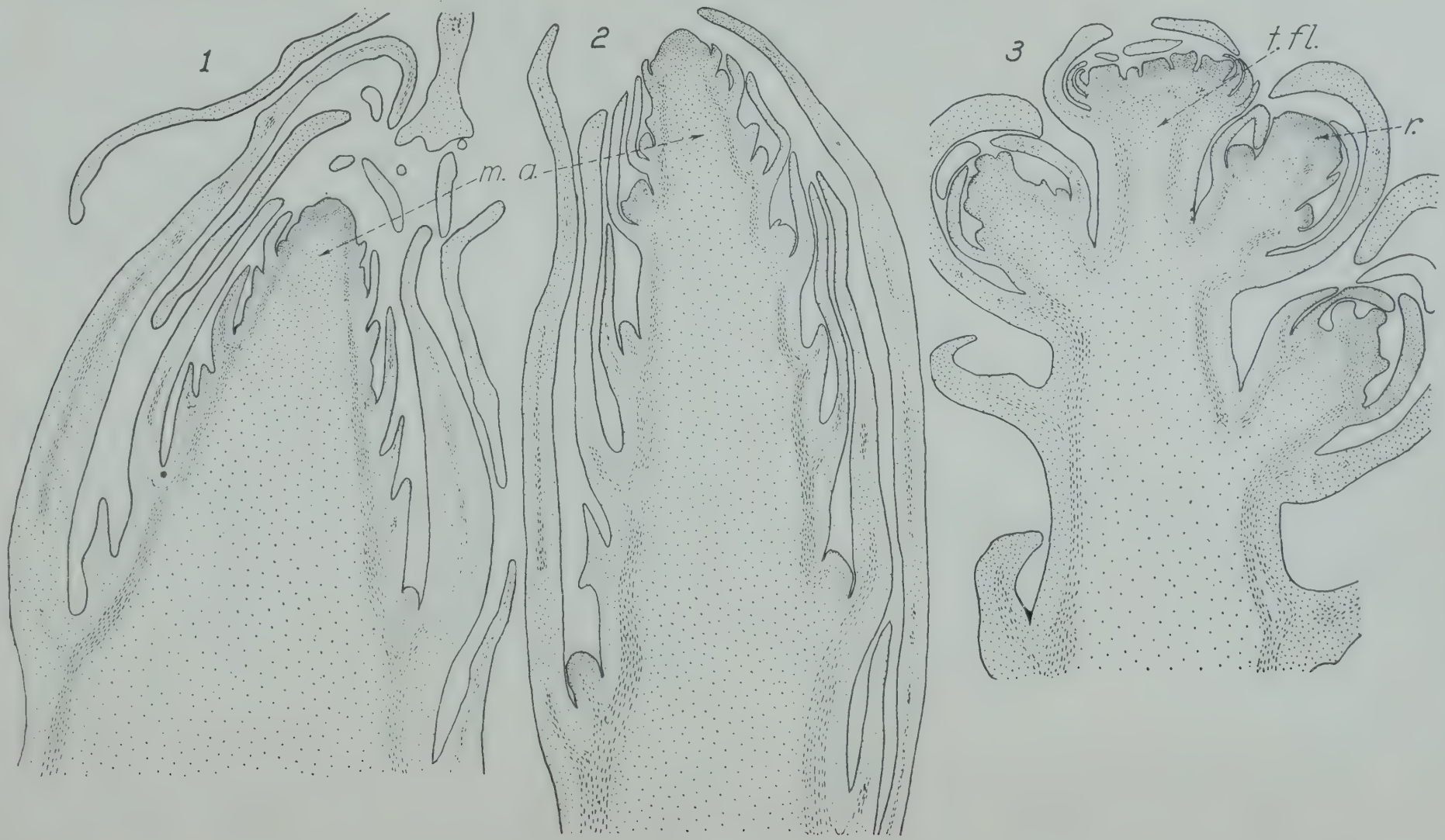
PLATE 1

All figures *ca.* × 30.

Fig. 1. Terminal portion of central axis. Heads soft, in best condition for quartering or removing upper leaves to allow the emergence of seed stalk.

Fig. 2. Terminal portion of central axis. Heads in prime condition for cutting for market.

Fig. 3. Terminal portion of central axis.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m. a.
2
3
t. fl.
r.

PLATE 2

Figs. 4 and 5. Receptacles, convex in figure 4 and flattened in figure 5. *Ca.* × 65.

Fig. 6. Flower head with protuberances that give rise to the primordia of the floral organs. *Ca.* × 65.

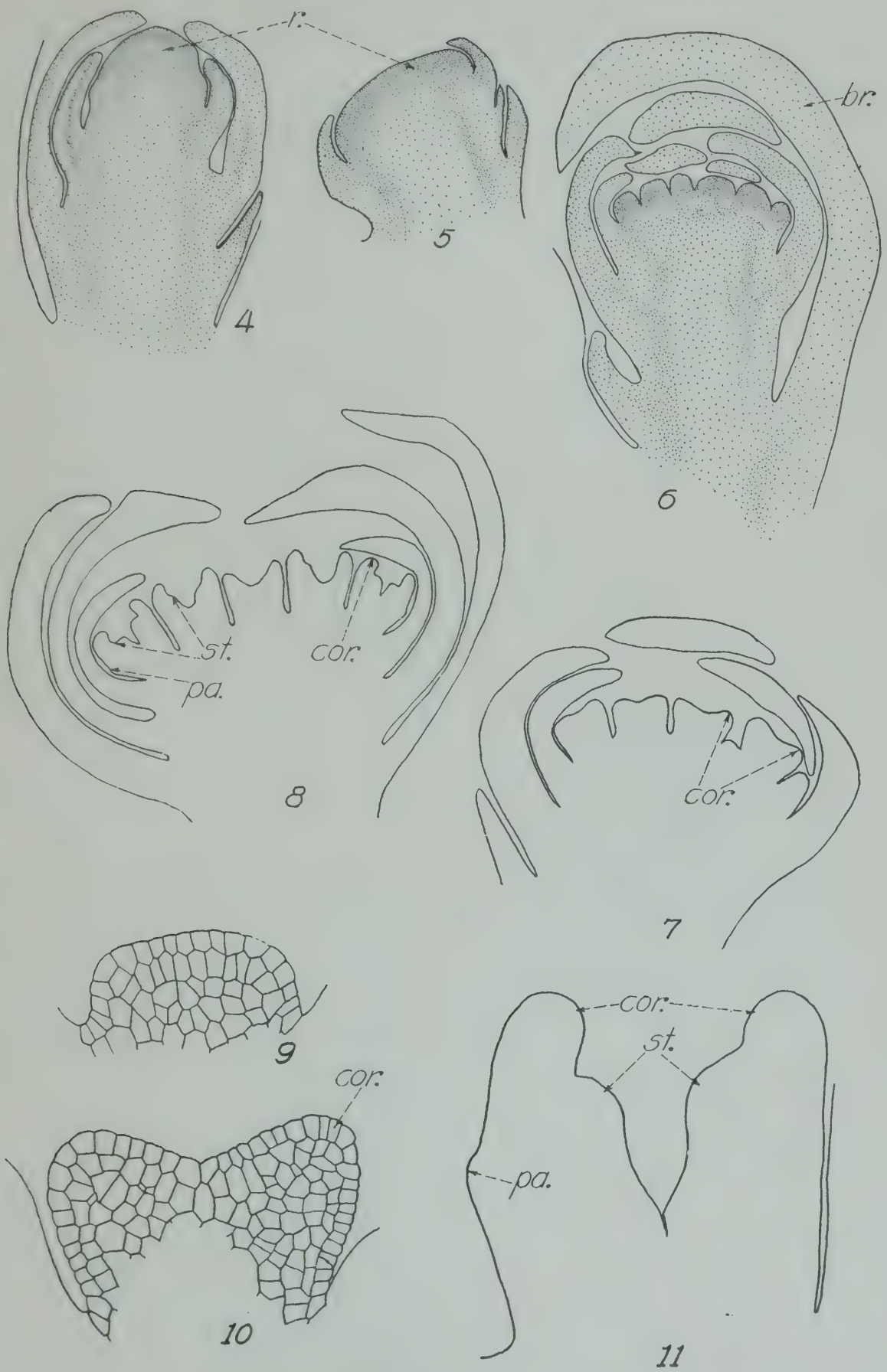
Fig. 7. Flower head showing the appearance of the corolla. *Ca.* × 65.

Fig. 8. Flower head, showing the corolla, stamens, and pappus. *Ca.* × 65.

Fig. 9. Detail of protuberance that later gives rise to flower primordia.

Fig. 10. Detail of early development of corolla.

Fig. 11. Individual flower. *Ca.* × 345.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br.
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cor.
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cor.
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cor.
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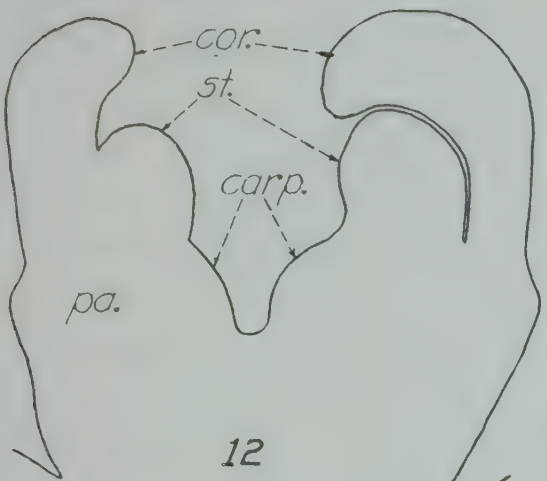
PLATE 3

Figs. 12 and 13. Development of floral organs. *Ca.* × 345.

Figs. 14 to 16. Developing ovule with megaspore mother cell, shaded. *Ca.* × 700.



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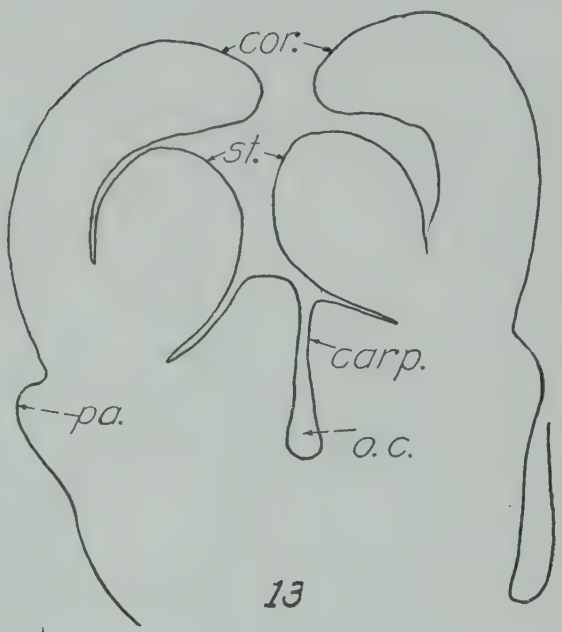

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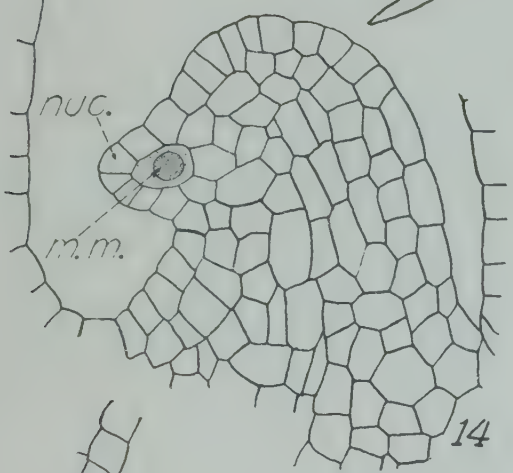

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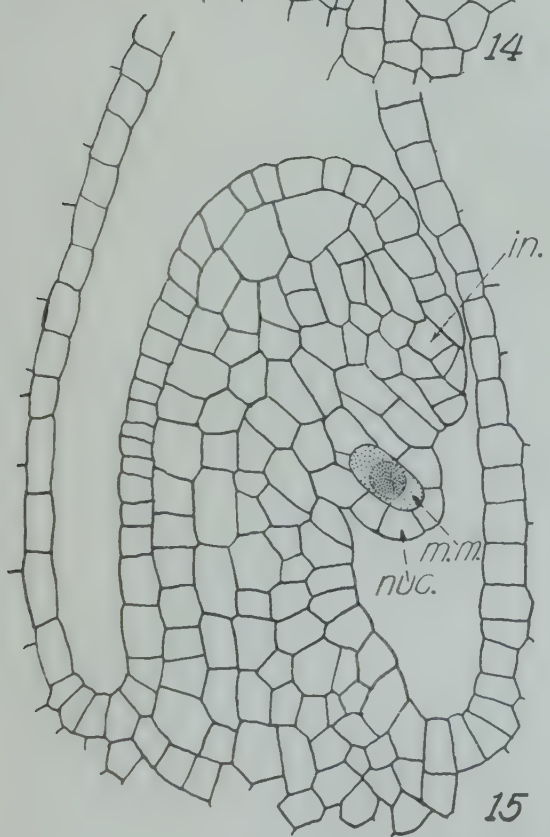

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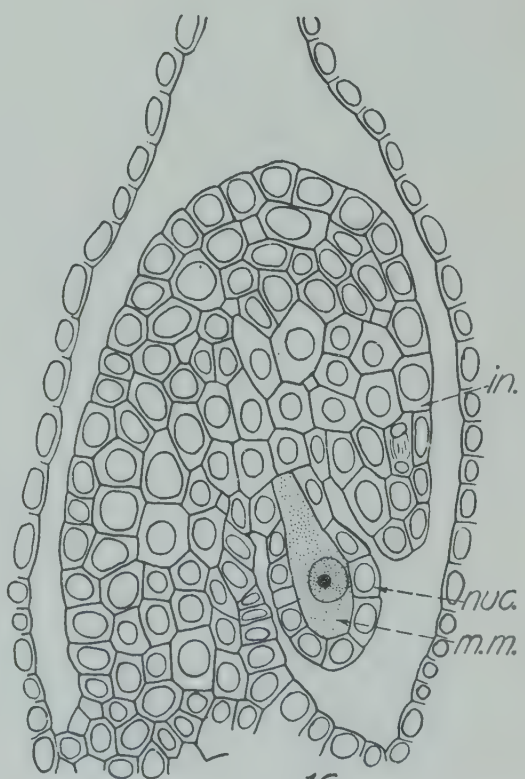

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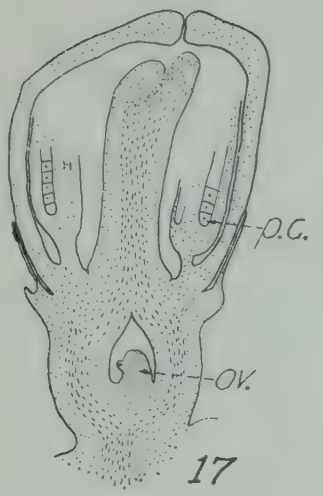
PLATE 4

Fig. 17. Longitudinal section through flower. Anthers in mother cell stage. Ovule in stage of development shown in figure 14. *Ca.* × 30.

Fig. 18. Transverse section through ovary and ovule. Megaspore mother cell and nucellus shaded. Megaspore mother cell in about the same stage of development as shown in figure 19. *Ca.* × 700.

Fig. 19. Ovule, showing large fleshy integument, the single layered nucellus, and the mature megaspore mother cell. *Ca.* × 700.



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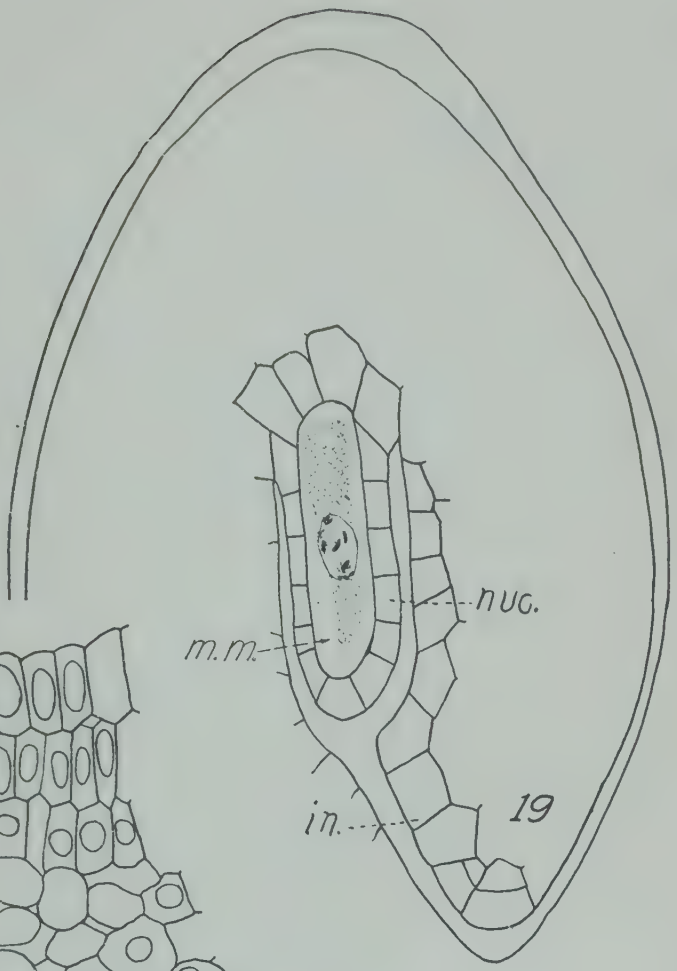

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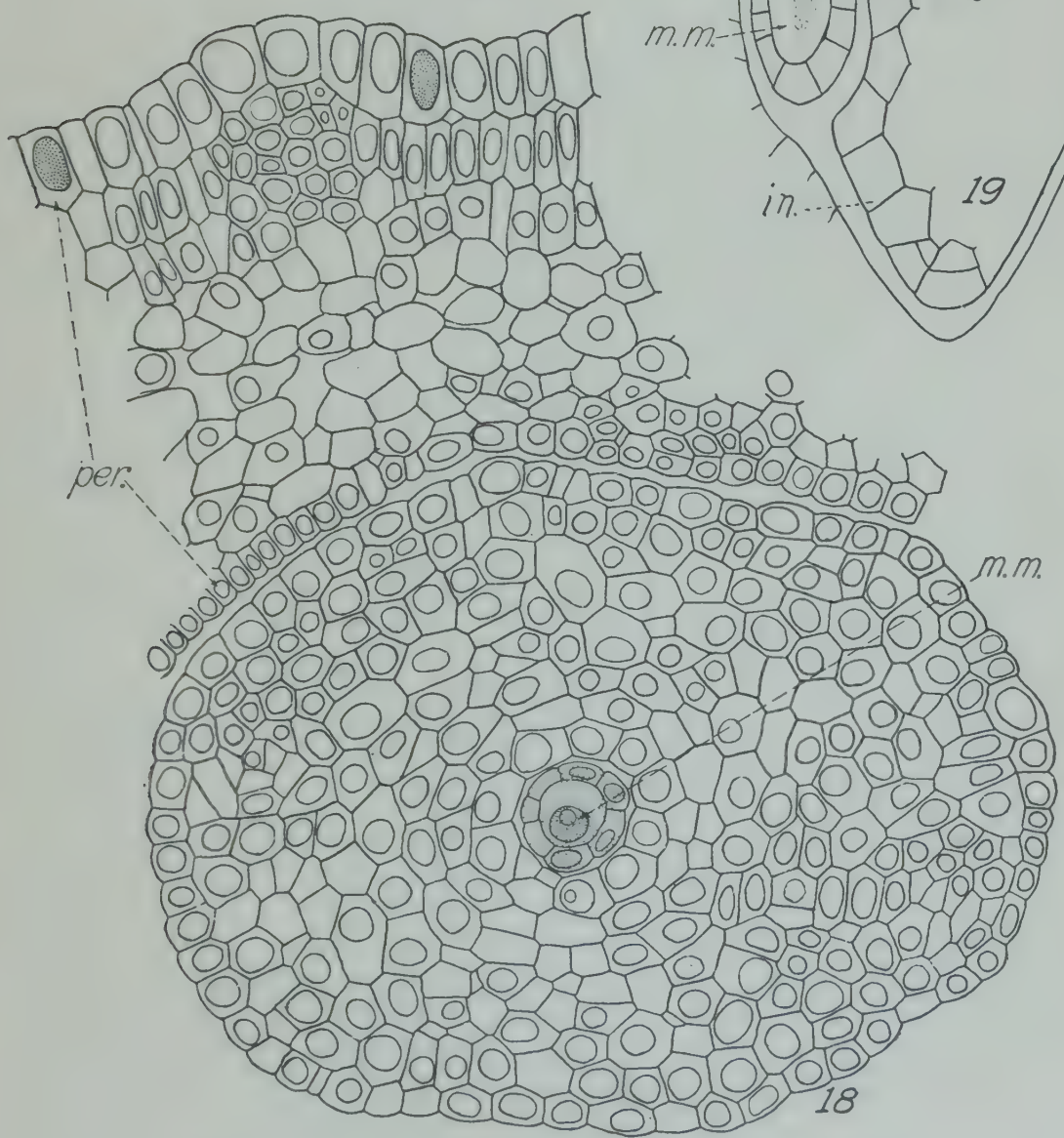

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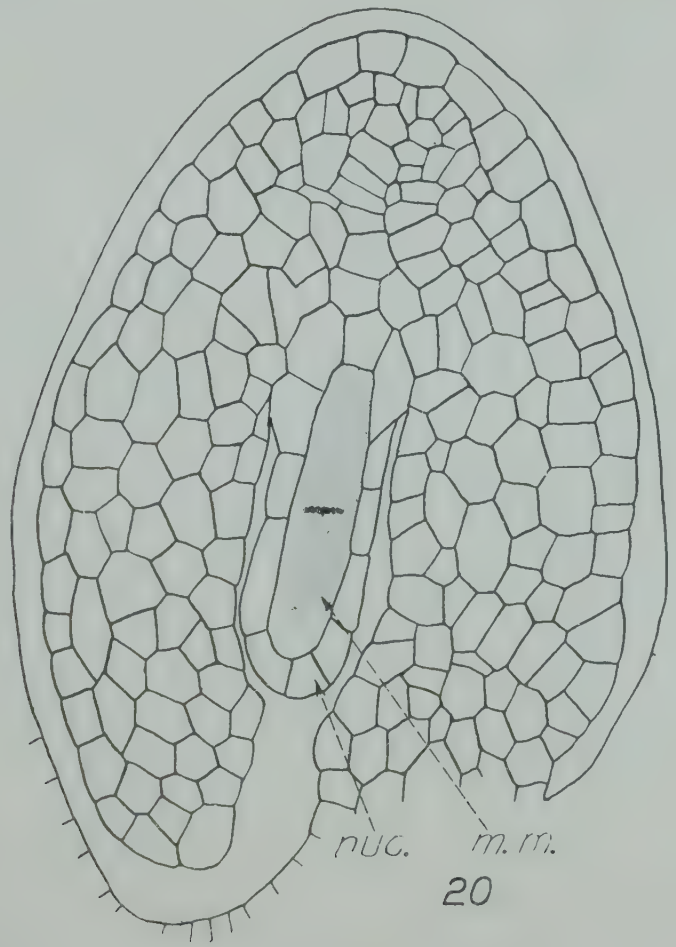
PLATE 5

All figures *ca.* × 700.

Fig. 20. Metaphase of heterotypic division of the megaspore mother cell.

Fig. 21. Metaphase of homotypic division of the daughter cells.



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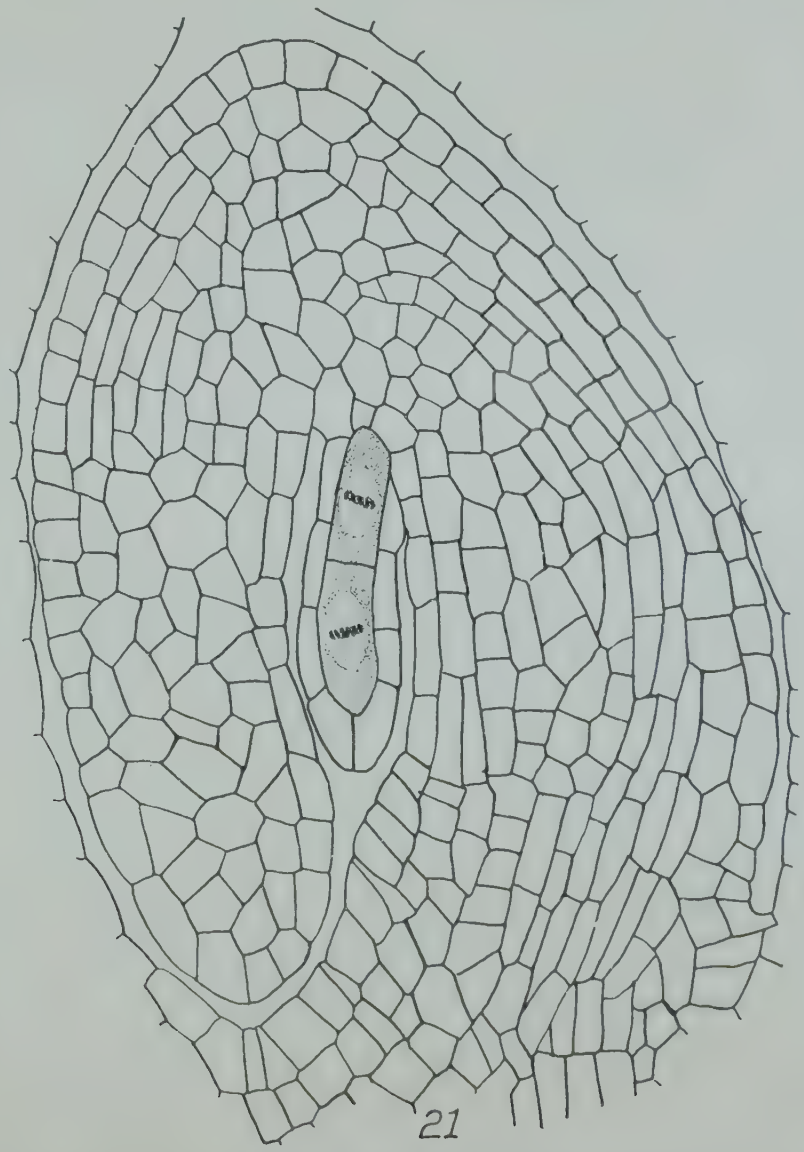

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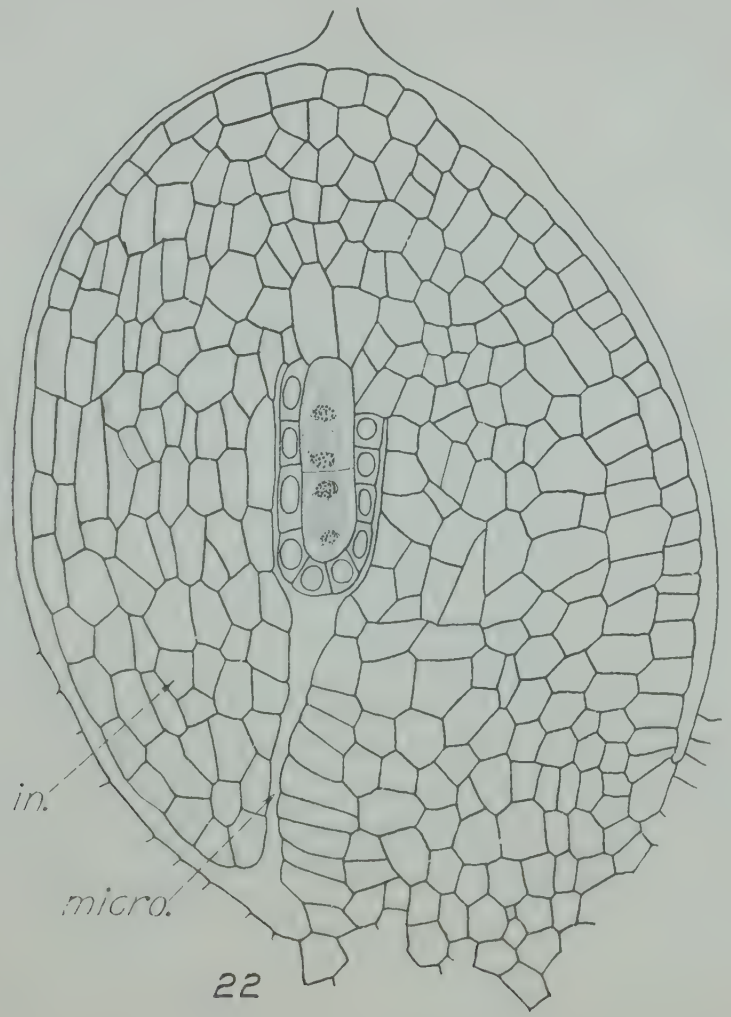
PLATE 6

All figures *ca.* × 700.

Fig. 22. Telophasic division of the two daughter cells.

Fig. 23. Tetrad of megaspores.



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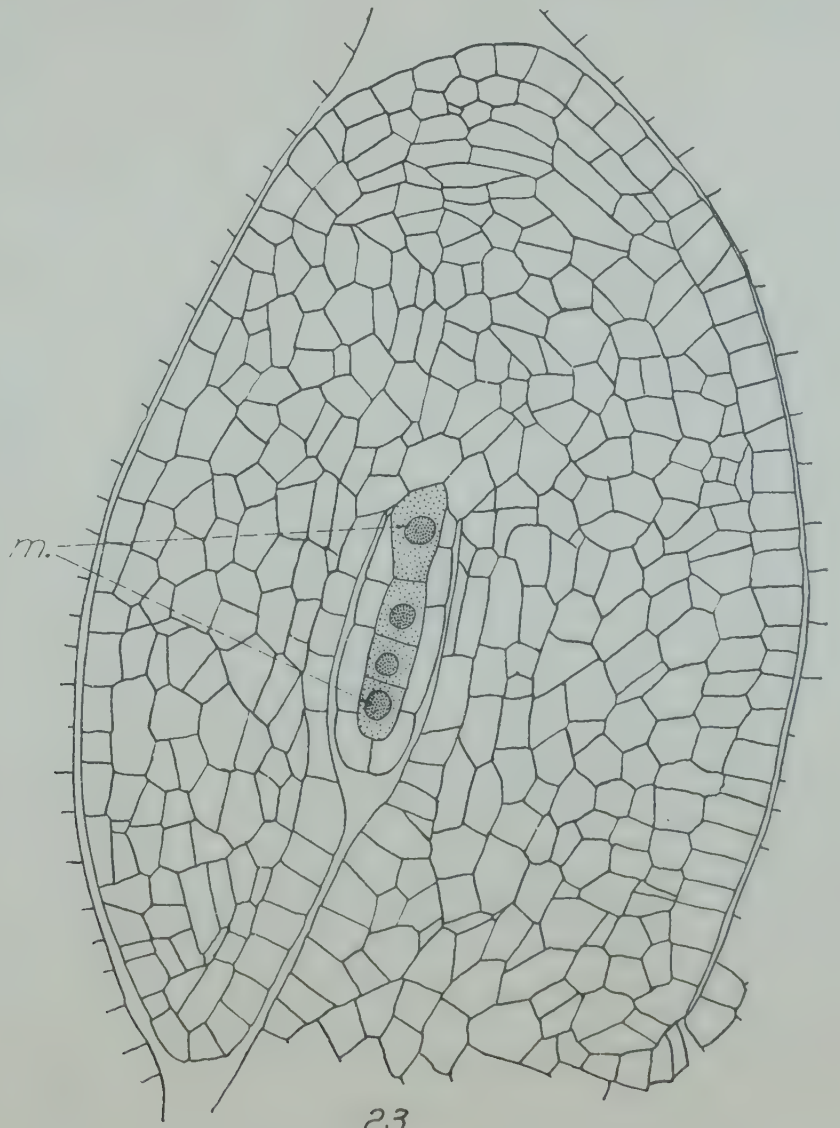

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PLATE 7

All figures *ca.* × 30.

Fig. 24. Transverse section through entire flower head. Anthers in pollen mother cell stage.

Fig. 25. Transverse section through flower head. Sample collected 5 A.M., June 12, 1924, three hours before pollination. Note the ovary with two ovules, one of which has two embryo sacs.

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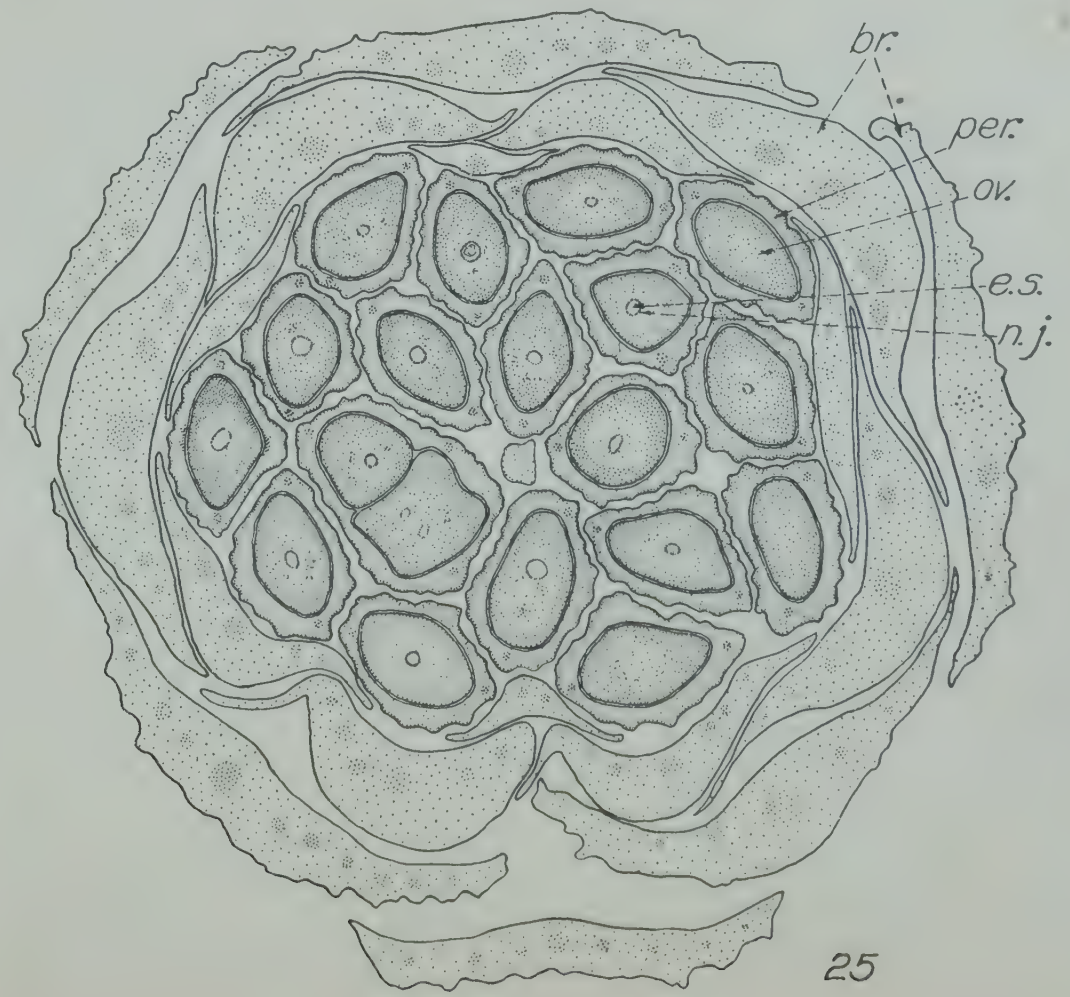

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PLATE 8

Fig. 26. Growth of the functioning megaspore and degeneration of the three lower (micropylar) megaspores. *Ca.* × 700.

Fig. 27. Embryo sac with two nuclei. *Ca.* × 700.

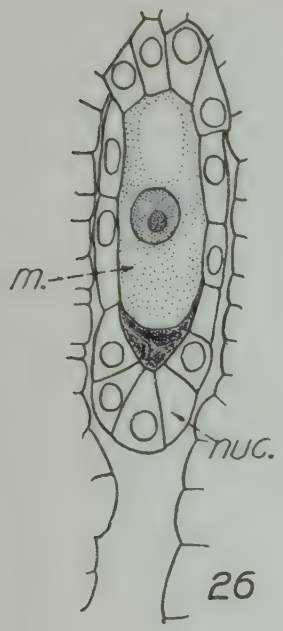
Fig. 28. Embryo sac with two nuclei, the latter migrating toward the poles. *Ca.* × 700.

Fig. 29. Longitudinal section of entire flower with ovule in same stage of development as shown in figure 28. The tetrads have escaped from the wall of the pollen mother cell. *Ca.* × 30.

Fig. 30. Embryo sac with four nuclei. *Ca.* × 700.

Fig. 31. Stigmatic papillae from inner surface of stigmatic lobes in stage of development shown in figure 29. *Ca.* × 700.

Fig. 32. Portion of mature embryo sac. *Ca.* × 700.



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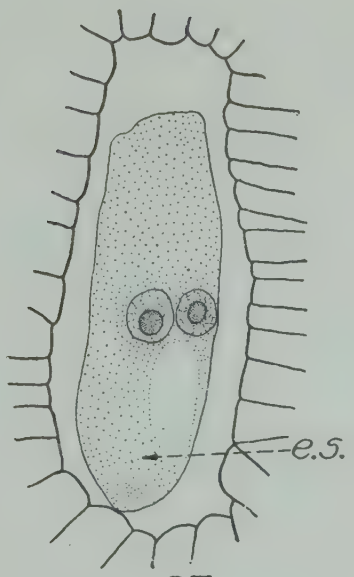

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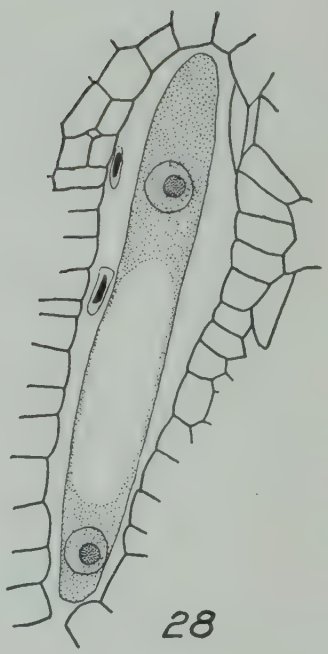

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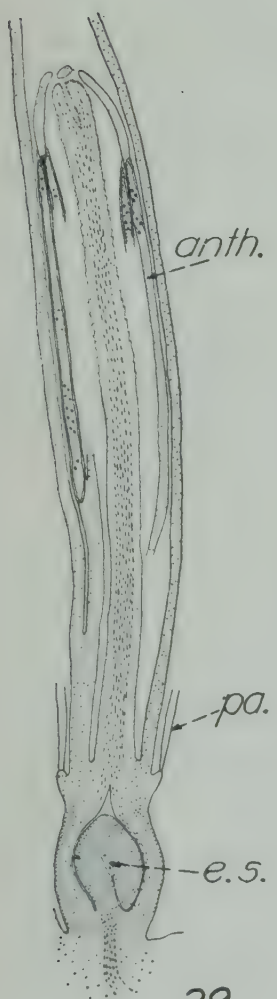

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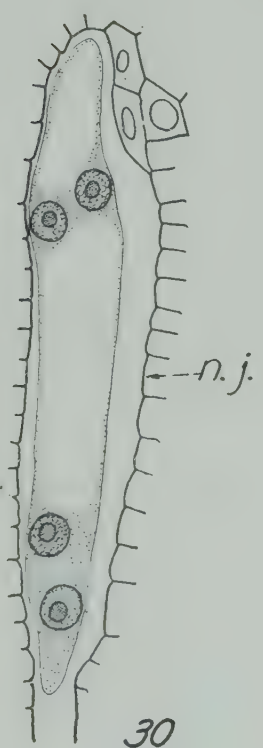

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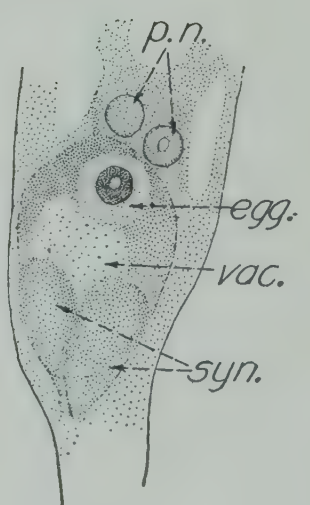

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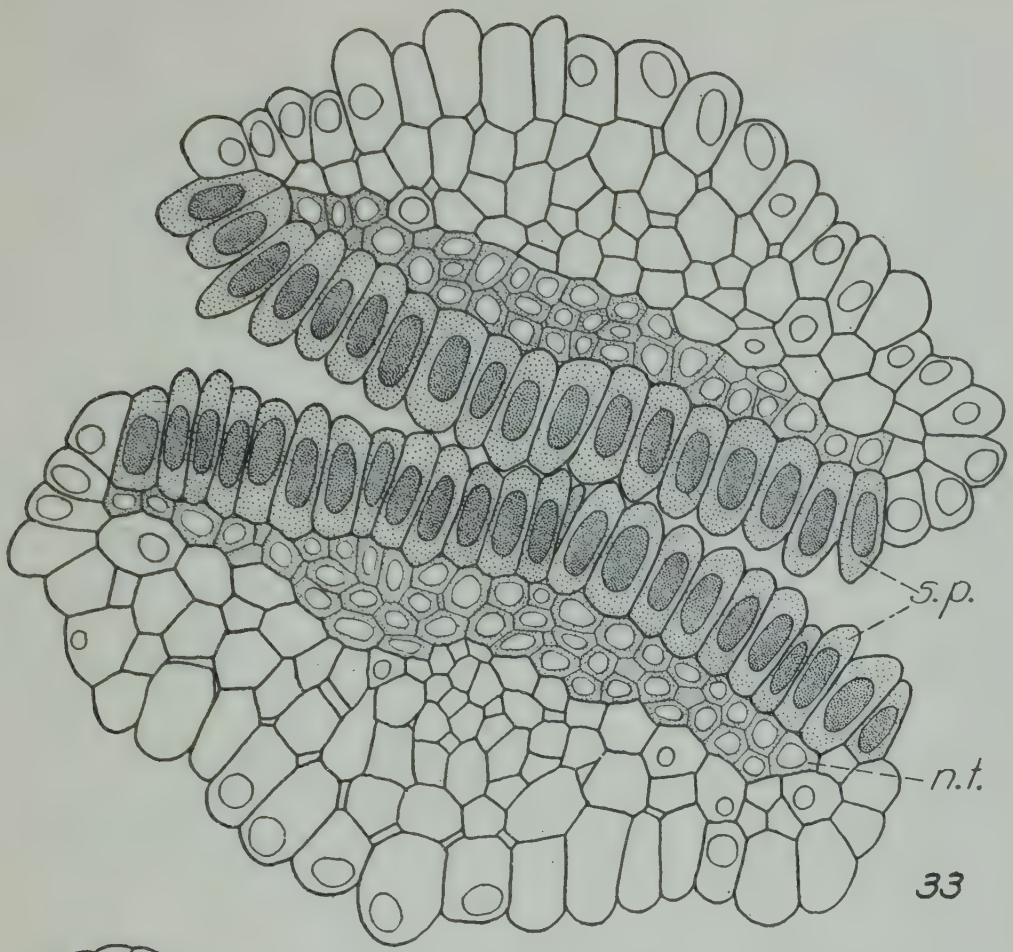
PLATE 9

All figures *ca.* × 700.

Fig. 33. Transverse section through stigmatic lobes. Stigmatic hairs develop from the deeply stained, elongated, large-nucleate cells.

Figs. 34 and 35. Longitudinal sections of young anthers showing single row of archesporial cells.

Fig. 36. Four archesporial cells have divided and cut off pollen mother cells on the inside.



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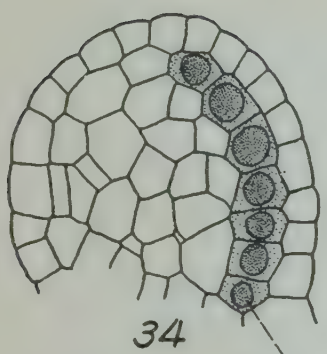

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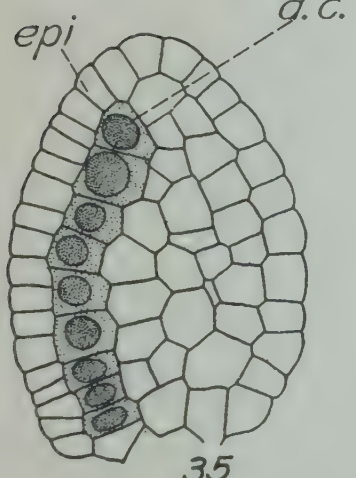

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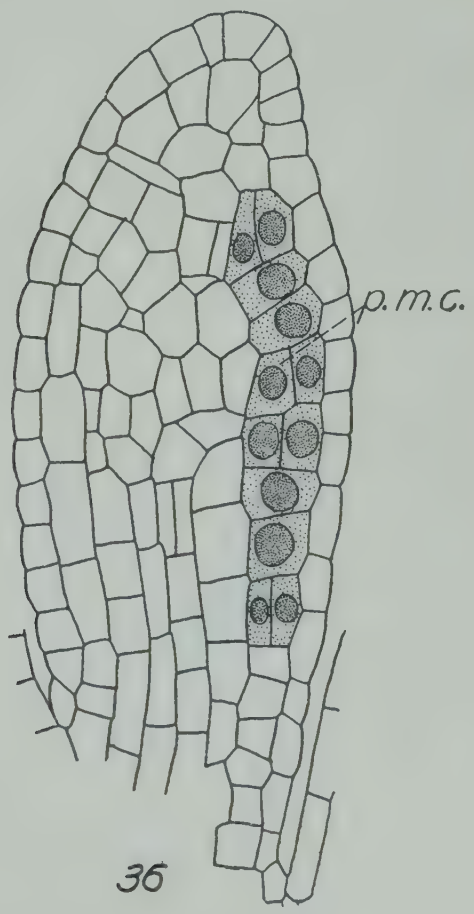

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PLATE 10

All figures *ca.* × 700.

Fig. 37. Single row of pollen mother cells and two-nucleate tapetal cells.

Fig. 38. Mother cells rounding off.

Fig. 39. Metaphase of heterotypic division of pollen mother cells. Four-nucleate tapetal cells.

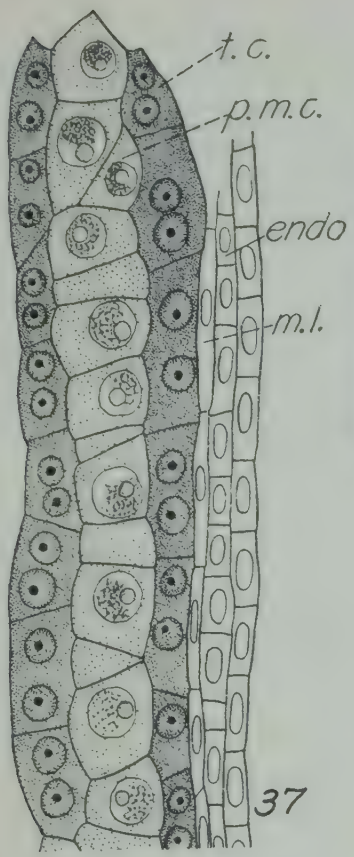
Fig. 40. Late anaphase of heterotypic division of pollen mother cells.

Fig. 41. Metaphase of homotypic division of the daughter cells.

Fig. 42. Transverse section of pollen sac. Tetrads enclosed within the wall of the pollen mother cell.

Fig. 43. Tetrads rounding off.

Fig. 44. Mature pollen grains, showing filamentous sperms and vegetative nucleus. Sample collected at 5 A.M., June 12, 1924, three hours before the time of pollination.



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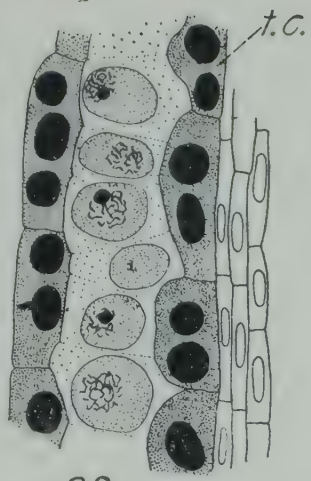

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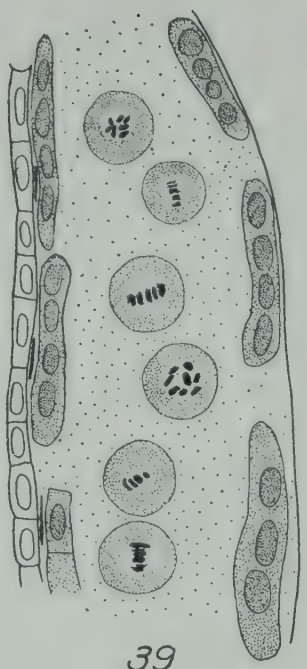
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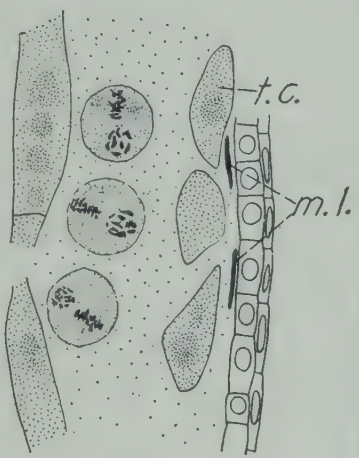

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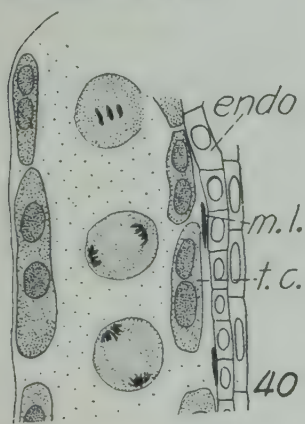

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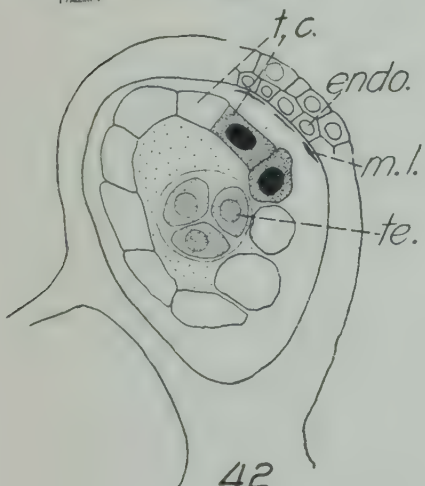

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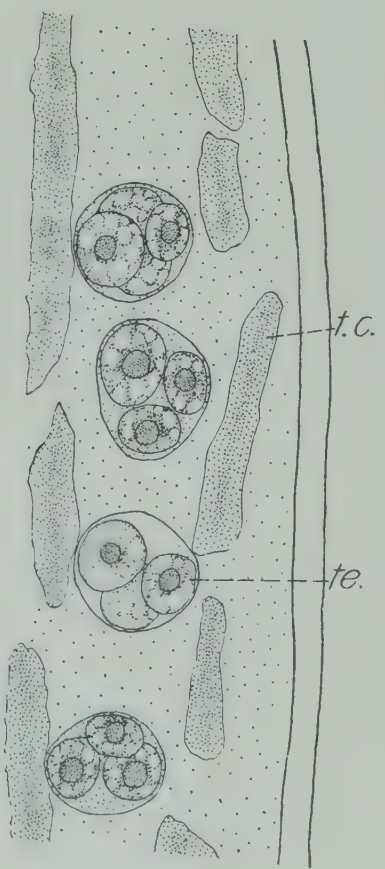

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PLATE 11

All figures *ca.* × 700.

Fig. 45. Fertilization: sperm nucleus within the egg cell; polar nuclei fusing. One P.M., June 12, 1924, five hours after pollination.

Fig. 46. Fertilization: one sperm nucleus in egg cell, the other in contact with the two polar nuclei, noon, June 12, 1924, four hours after pollination.

Fig. 47. Fertilized egg. Two P.M., June 12, 1924, six hours after pollination.

Fig. 48. First division of the zygote: metaphase. Two P.M., June 12, 1924, six hours after pollination.

Fig. 49. First division of fertilized egg: telophase. Two P.M., June 12, 1924, six hours after pollination.

Fig. 50. Two-celled embryo: *a*, upper; *b*, lower.

Fig. 51. Two-celled embryo: cell *a* and cell *b* in late anaphase. Five P.M., June 12, 1924, nine hours after pollination.

Fig. 52. Two-celled embryo: cell *a* in telophase and cell *b* in metaphase. Five P.M., June 12, 1924, nine hours after pollination.

Fig. 53. Cell wall being formed in cell *a;* cell *b* in metaphase. Five P.M., June 12, 1924, nine hours after pollination.

Fig. 54. Cell *a* in telophase; cell *b* in anaphase. Five P.M., June 12, 1924, nine hours after pollination.

Fig. 55. Three-celled embryo: tier *a* two-celled; cell *b* in telophase. Five P.M., June 12, 1924, nine hours after pollination.

Fig. 56. Four-celled embryo: *c*, and *d*, daughter cells of *b*. Six P.M., June 12, 1924, ten hours after pollination.

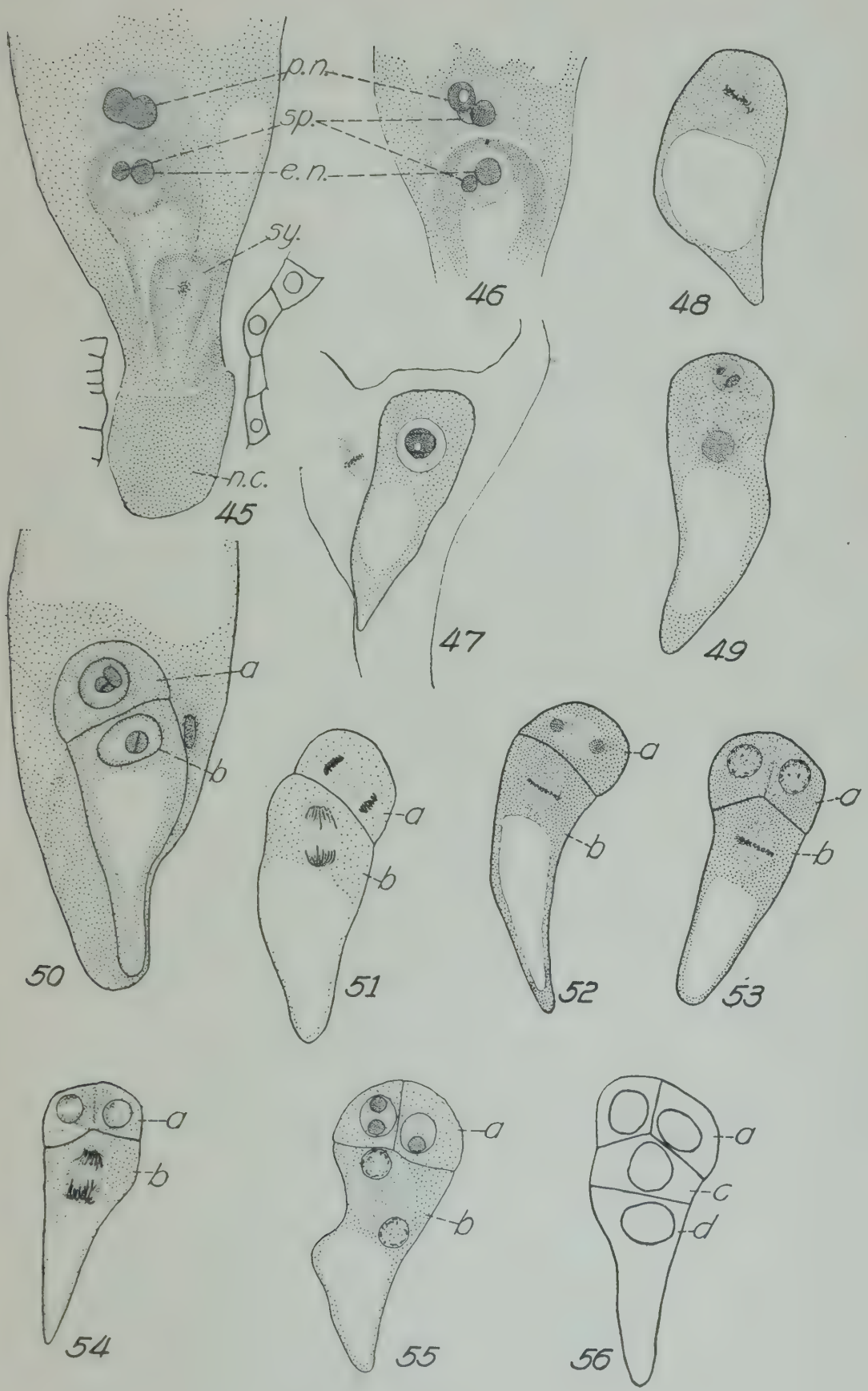
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PLATE 12

All figures *ca.* × 700.

Fig. 57. Four-celled embryo: transverse section through tier *a*. Nuclei of both cells in prophase; one of the endosperm nuclei in anaphase. Eight P.M., June 12, 1924, twelve hours after pollination.

Fig. 58. Four-celled embryo: both cells of tier *a* in prophase, cell *c* in metaphase, and cell *d* in prophase. Eight P.M., June 12, 1924, twelve hours after pollination.

Fig. 59. Four-celled embryo: both cells of tier *a* in anaphase, one slightly in advance of the other; cells *c* and *d* in prophase. Eight P.M., June 12, 1924, twelve hours after pollination.

Fig. 60. Four-celled embryo: both cells of tier *a* in metaphase, cell *c* in anaphase, and cell *d* in prophase. Eight P.M., June 12, 1924, twelve hours after pollination.

Fig. 61. Four-celled embryo: one cell of tier *a* in late anaphase, the other in metaphase; cell *c* in anaphase; and cell *d* in prophase. Ten P.M., June 12, 1924, fourteen hours after pollination.

Fig. 62. Antipodals. Eight P.M., twelve hours after pollination.

Fig. 63. Eight-celled embryo: tier *a*, four-celled; tier *c*, two-celled; *e* and *f*, daughter cells of *d*. Four A.M., June 13, 1924, twenty hours after pollination.

Fig. 64. Twelve-celled embryo: tier *a*, eight-celled; both cells of tier *c* in anaphase; cell *e* in prophase. Ten A.M., June 13, 1924, twenty-six hours after pollination.

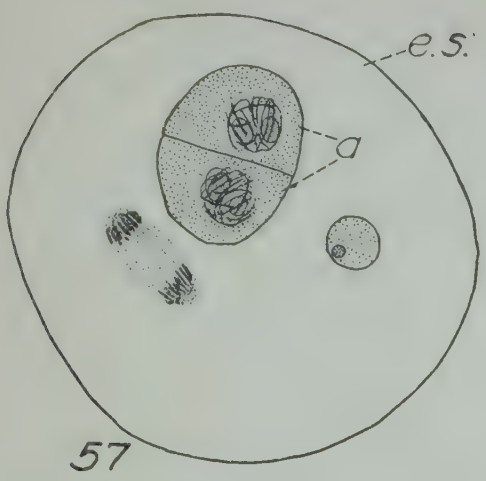
Fig. 65. Fourteen-celled embryo: tier *a* eight-celled; tier *c* four-celled; cell *e* in metaphase. Ten A.M., June 13, 1924, twenty-six hours after pollination.

Figs. 66 and 67. Fifteen-celled embryo: tier *a* eight-celled; tier *c*, four-celled; tier *e*, two-celled. Ten A.M., June 13, 1924, twenty-six hours after pollination.

Fig. 68. Fifteen-celled embryo: cell *f* in prophase. Ten A.M., June 13, 1924, twenty-six hours after pollination.

Figs. 69 and 70. Fifteen-celled embryos: cell *f* in metaphase. Ten A.M., June 13, 1924, twenty-six hours after pollination.

Fig. 71. Fifteen-celled embryo: cell *f* in telophase. Ten A.M., June 13, 1924, twenty-six hours after pollination.



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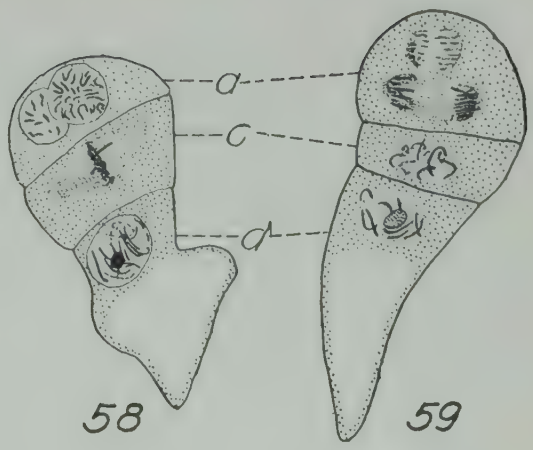

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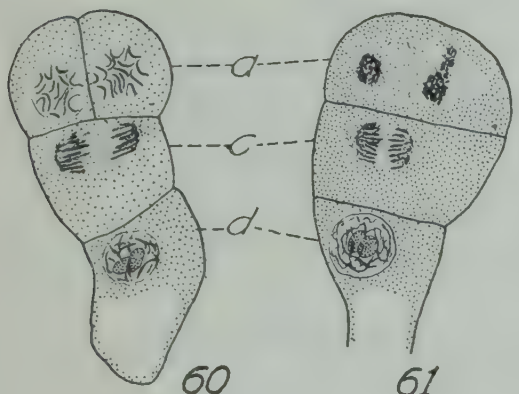

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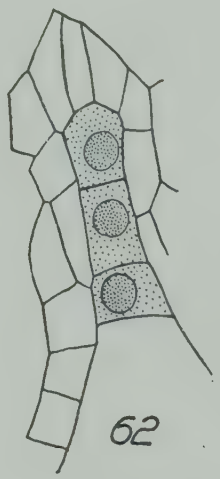
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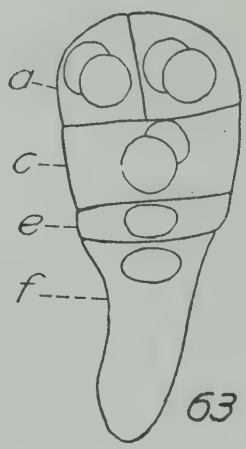

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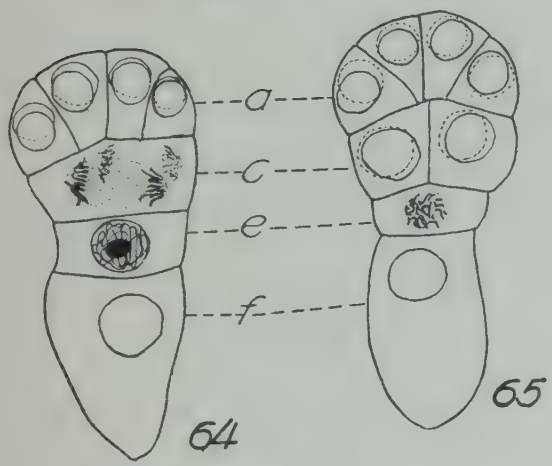

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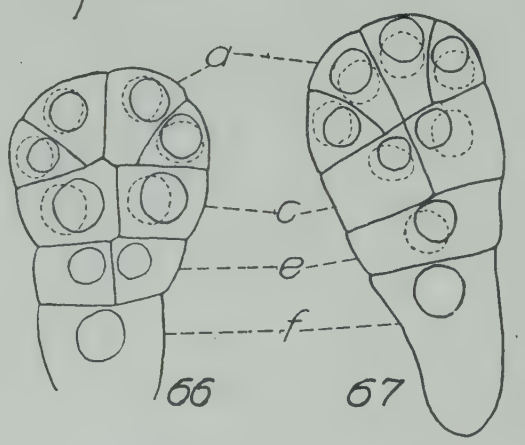

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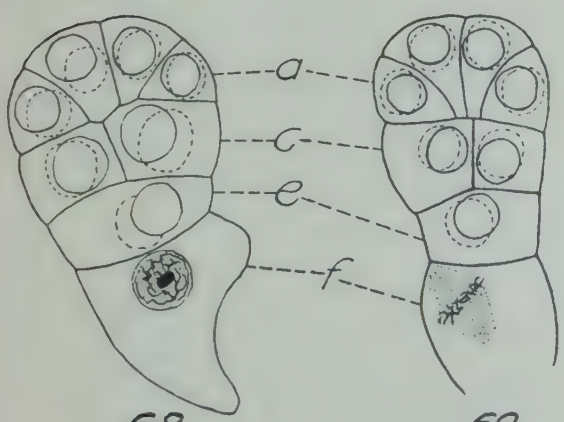

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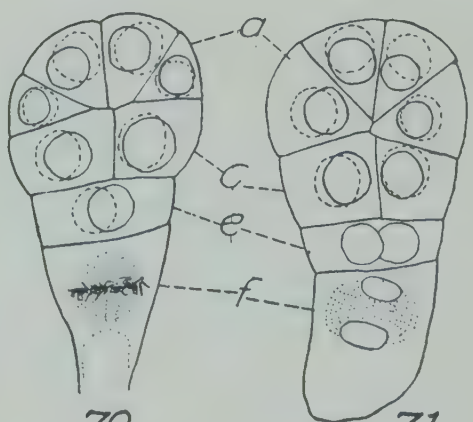

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PLATE 13

All figures *ca.* × 700.

Fig. 72. Sixteen-celled embryo: tier *a*, eight-celled; tier *c*, four-celled; tier *e*, two-celled; *g* and *h*, daughter cells of *f*. Ten A.M., June 13, 1924, twenty-six hours after pollination.

Figs. 73 and 74. In tiers *a* and *c*, dermatogen cells have been cut off; in tier *c*, periblem and plerome cells have been differentiated. Six P.M., June 13, 1924, thirty-four hours after pollination.

Fig. 75. One cell of the plerome in tier *c* has divided transversely. Dermatogen cells and periblem initials have been differentiated in tier *e*. Six A.M., June 14, 1924, forty-six hours after pollination.

Fig. 76. Both plerome cells of tier *c* have been divided into two by transverse walls. Six A.M., June 14, 1924, forty-six hours after pollination.

Fig. 77. Tier *c* has become a 3- to 4-celled layer. Eight A.M., June 15, 1924, three days after pollination.

Fig. 78. Cotyledonary swellings appearing. Eight A.M., June 15, 1924, three days after pollination.

Fig. 79. Lower portion of embryo. Cells of root cap (shaded); dermatogen cells with nuclei shown. Eight A.M., June 16, 1924, four days after pollination.

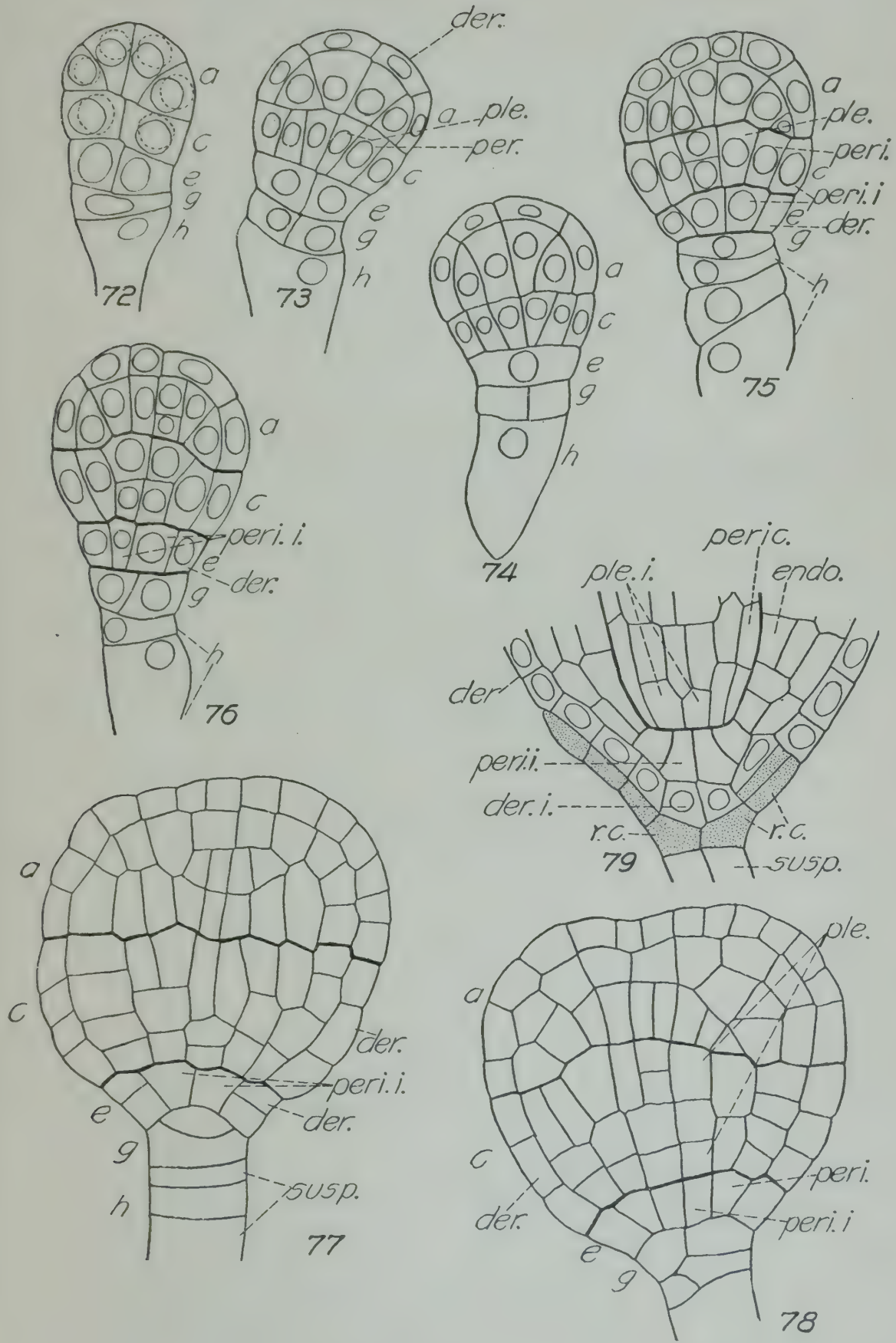
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PLATE 14

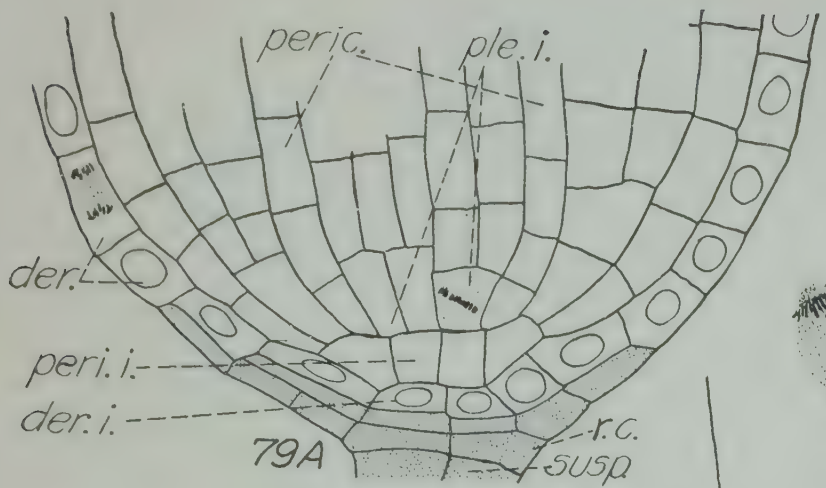
All figures *ca.* × 700.

Fig. 79*a*. Lower portion of embryo. Cells of root cap (shaded), and dermatogen cells with nuclei shown. Eight A.M., June 16, 1924, four days after pollination.

Fig. 80. Four-celled embryo: fourth division of the free endosperm nuclei. Eight P.M., June 12, 1924, twelve hours after pollination.

Fig. 81. Eight-celled embryo: walled endosperm cells. Four A.M., June 13, 1924, twenty hours after pollination.

Fig. 82. Longitudinal section of embryo sac showing endosperm tissue just above the developing embryo. Eight A.M., June 15, 1924, three days after pollination.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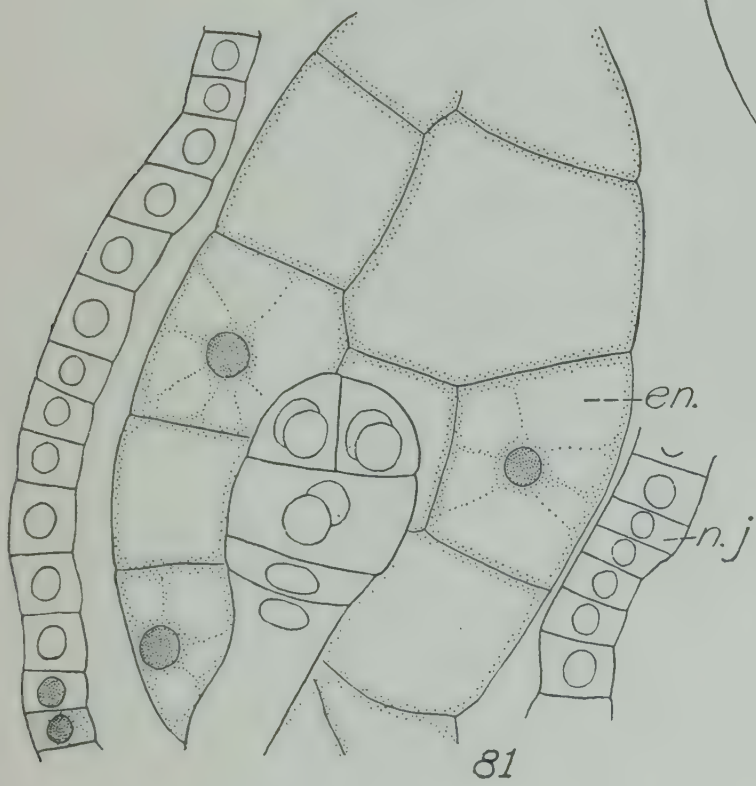
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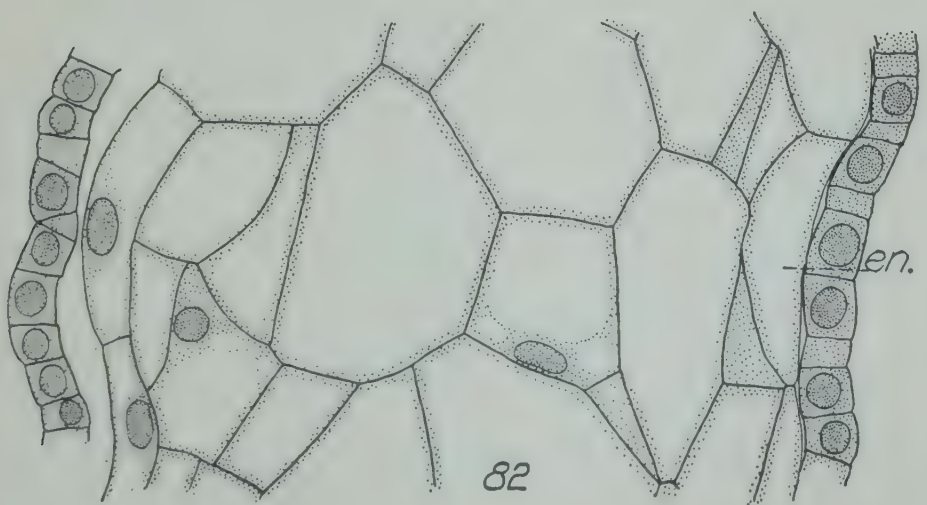
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PLATE 15

All figures *ca.* × 5.

Fig. 83. Flower head twenty-four hours preceding anthesis.

Fig. 84. Single bud of head shown in figure 83.

Fig. 85. Flower head two hours preceding anthesis.

Fig. 86. Single flower, just before full bloom. Pollen-covered pistil not fully extended.

Fig. 87. Single flower in full bloom. Pistil is fully extended and covered with pollen.

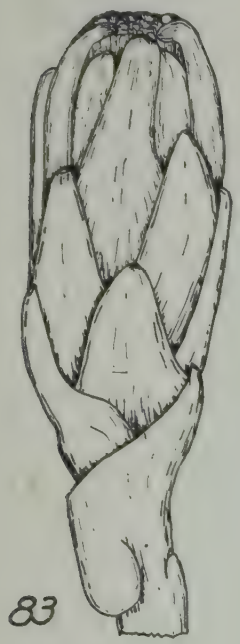
Fig. 88. Twenty-four hours after anthesis. Withered corollas, stamens, and styles still attached.

Fig. 89. Individual flower from head shown in figure 88.

Fig. 90. Developing achenes; forty-eight hours after pollination.

Fig. 91. Seed head three days after pollination.

Fig. 92. Developing achenes from head shown in figure 91.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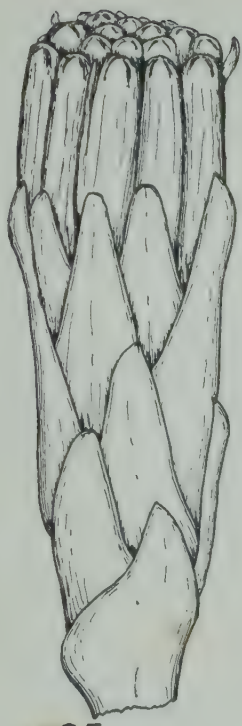
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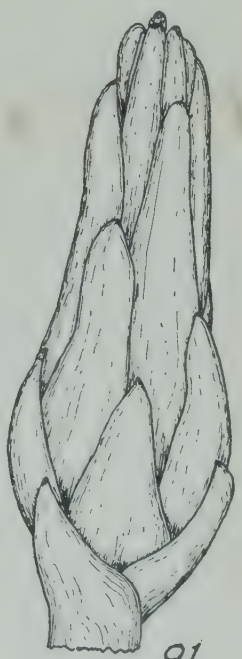
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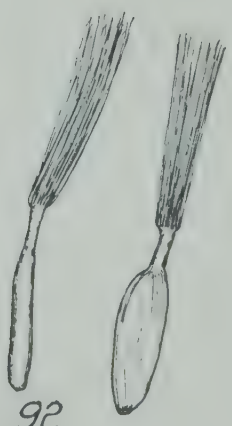
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PLATE 16

Figs 93 to 95 *ca.* × 5; figs. 96 to 102 *ca.* × 30.

Fig. 93. Developing achene four days after pollination.

Fig. 94. Seed head five days after pollination.

Fig. 95. Developing achene six days after pollination.

Figs. 96 to 102. Longitudinal sections through the developing fruit.

Fig. 96. Six P.M., June 12, 1924, ten hours after pollination.

Fig. 97. Ten P.M., June 13, 1924, fourteen hours after pollination.

Fig. 98. Ten A.M., June 13, 1924, twenty-six hours after pollination.

Fig. 99. Six P.M., June 13, 1924, thirty-four hours after pollination.

Fig. 100. Six A.M., June 14, 1924, forty-six hours after pollination.

Fig. 101. Eight A.M., June 16, 1924, four days after pollination.

Fig. 102. Eight A.M., June 17, 1924, five days after pollination.

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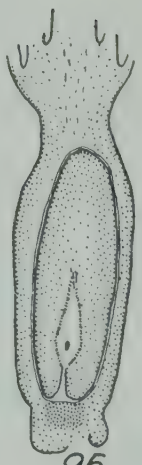
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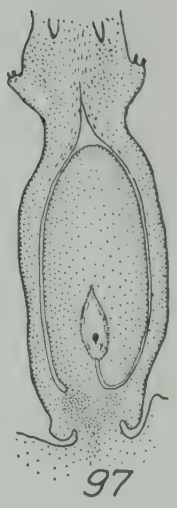
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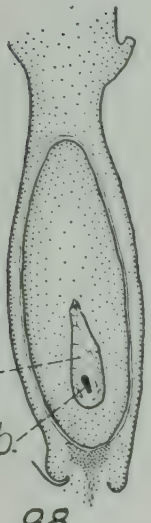

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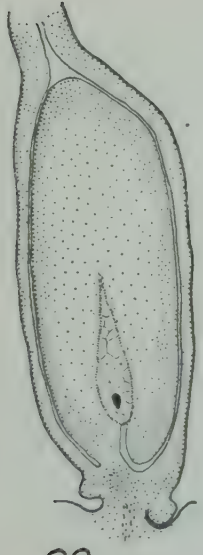
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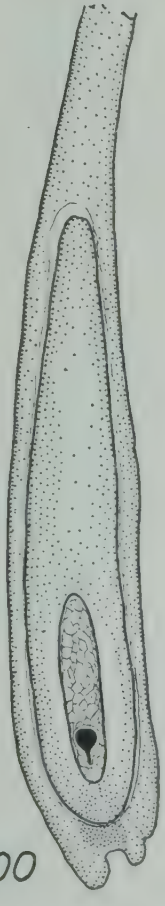
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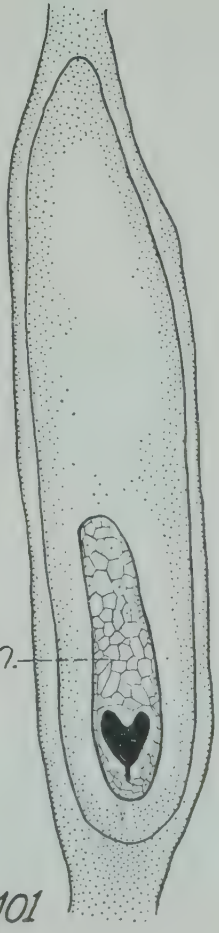

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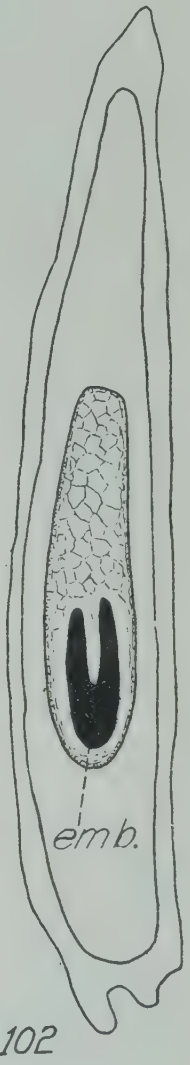

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PLATE 17

All figures *ca.* × 30.

Fig. 103. Eight A.M., June 18, 1924, six days after pollination.

Fig. 104. Eight A.M., June 19, 1924, seven days after pollination.

Fig. 105. Seven A.M., June 20, 1924, eight days after pollination.

Fig. 106. Eight A.M., June 23, 1924, eleven days after pollination. (The following day the achenes of the same age were ripe.)

Fig. 107. Transverse section through developing achenes. Eight A.M., June 21, 1924, nine days after pollination. *A*, radicle; *B*, cotyledons and plumule; *C*, cotyledons.

Fig. 108. Transverse section through developing achene. Eight A.M., June 23, 1924, eleven days after pollination. *A*, radicle; *B*, cotyledons and plumule; *C*, cotyledons.

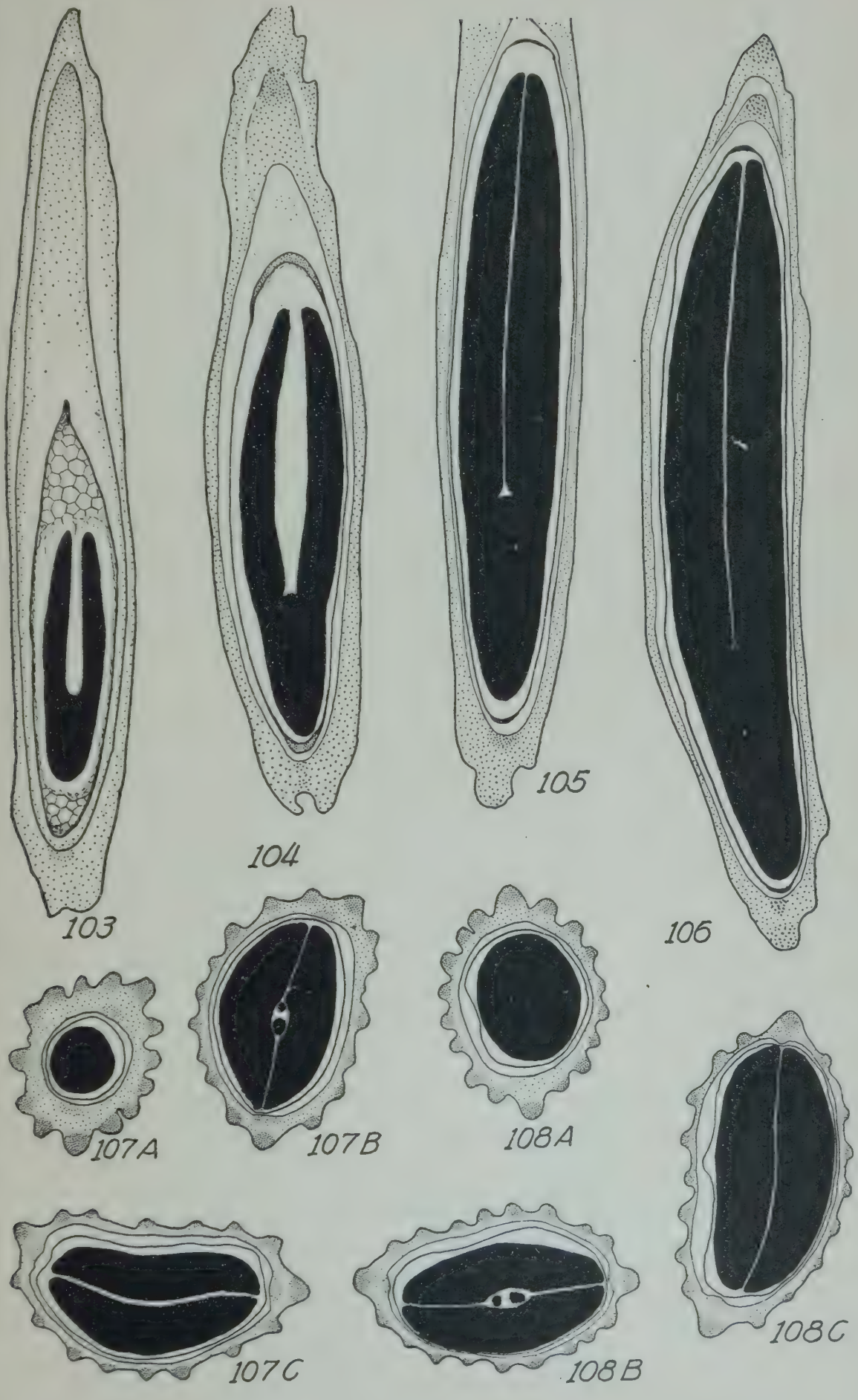
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PLATE 18

Fig. 109. Flowering and seed ripening curves for plant No. 8. Mean temperature is given in degrees Fahrenheit.

Fig. 110. Flowering and seed ripening curves. Average of all plants.

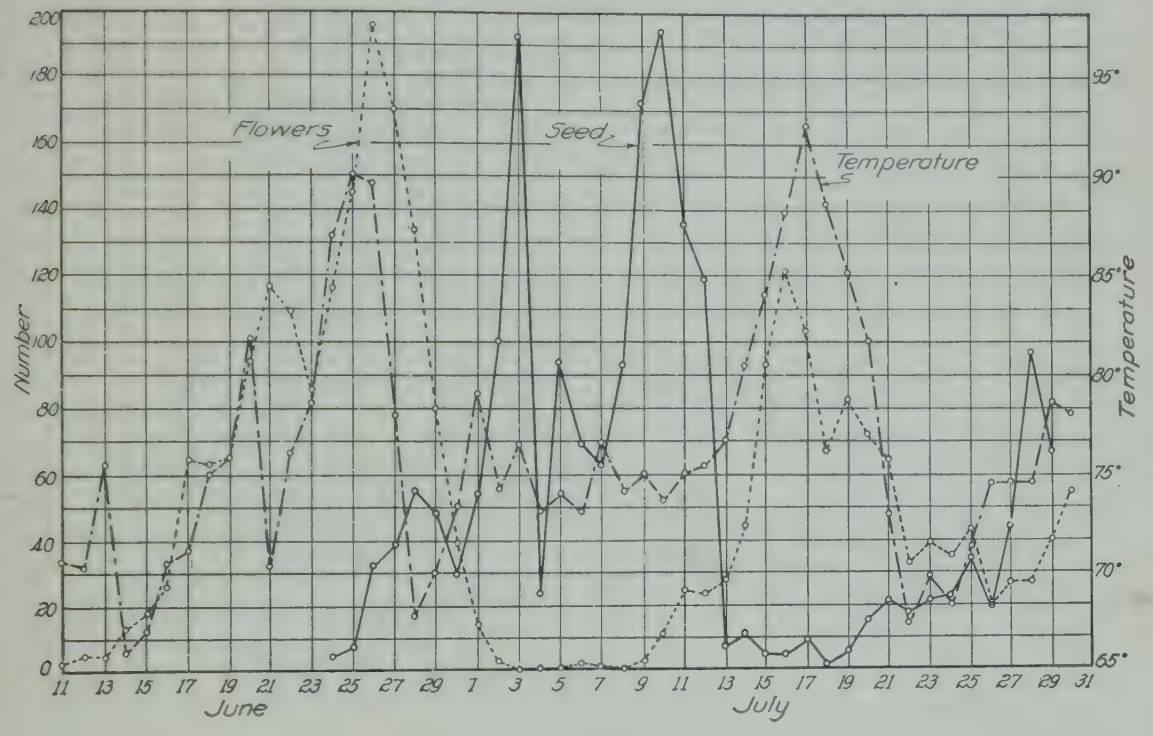

Fig. 109

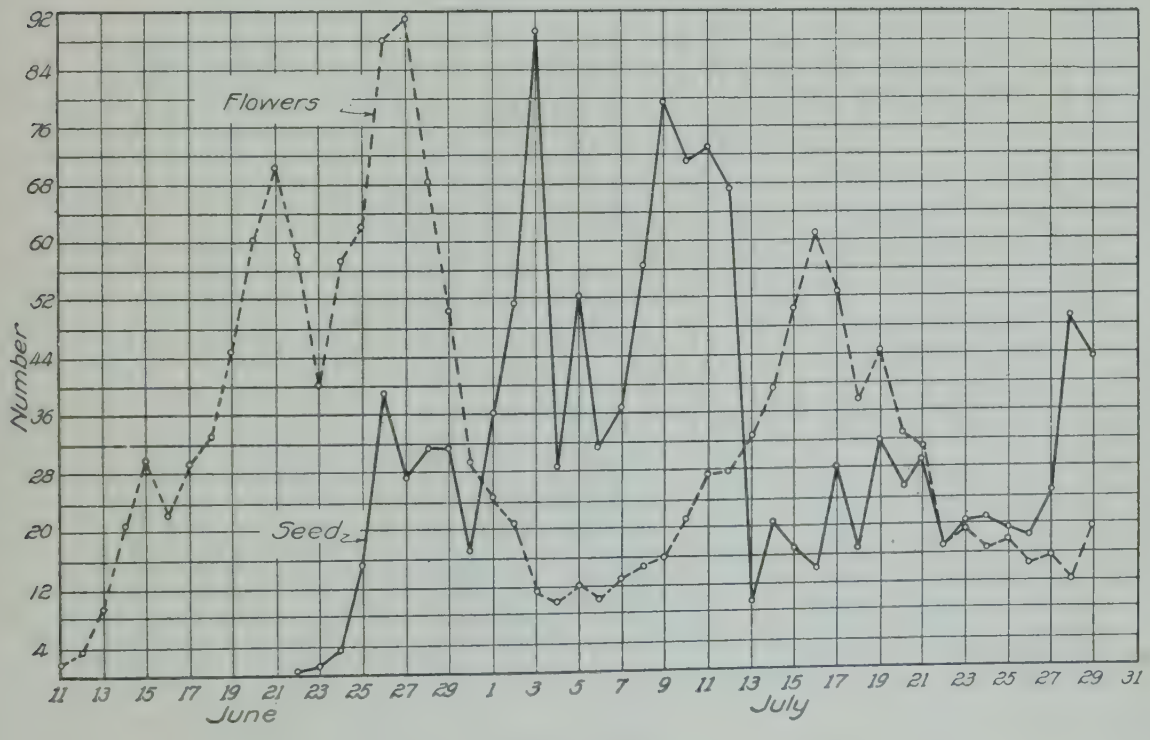

Fig. 110

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THE APPLICATION OF HYDRODYNAMICS TO IRRIGATION AND DRAINAGE PROBLEMS*

ORSON W. ISRAELSEN

The maintenance of soil fertility in irrigated regions depends in part on control of the movement of ground water† and of soil moisture. It is apparent that knowledge of the laws governing the flow of water in soils is essential to the effective control of its movement. Much experimental work has been done on the movement of water in soils but comparatively little use has been made of the fundamental laws of motion, either as a guide to experimental procedure, or in the interpretation of the experimental observations. Intelligent guidance to irrigation engineers, to managers of irrigation systems, and indeed to practical irrigators, in the proper use of irrigation water and in the solution of problems in the maintenance of soil fertility which arise from its improper use, is dependent on a knowledge of the laws which control the movement of water in soil. It is, therefore, important that experimenters studying movement of moisture in soils should ascertain to what extent the fundamental laws of motion of fluids advantageously may be applied to their problems.

Hydrodynamics is that branch of physics which deals with the motion of fluids. The term fluids includes both liquids and gases, but

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* A thesis submitted to satisfy in part the requirements of the University of California for the degree of Doctor of Philosophy.

† The term ground water as here used refers to that water which completely fills the pore spaces in the soil; whereas the term soil moisture signifies the water which exists in the capillary form and which does not ordinarily completely fill all of the soil pore space.

the purposes of this paper require only a study of liquids. The science of hydrodynamics is essentially mathematical. To simplify the mathematics, it is customary in the preliminary analysis to assume that the fluids dealt with are frictionless and hence that they support only normal stresses. The fluids are also assumed to be continuous throughout the space considered.

In preparing this paper the writer has realized that some of his readers may find difficulty in following the mathematical methods employed. It is hoped, however, that those who have some knowledge of the elementary calculus will be able to understand the analysis. The most general case of the dynamics of deformable bodies involves tedious analytical difficulties, a consideration of which the purposes of this paper do not require. The basic courses in mechanics for engineers as usually taught are concerned primarily with rigid or semirigid bodies, and the college courses in hydraulics usually include only the applications of a few elementary principles of hydrodynamics. The law of conservation of energy is used in the development of Bernoulli's Theorem and this furnishes the primary hydrodynamical background in hydraulics.

The equation of continuity, although extensively employed in the solution of problems in hydraulics, is seldom either written or derived in its general form. Since hydraulics deals largely with non-compressible liquids and with one-dimensional flow, the use of Bernoulli's Theorem as the basis for the equation of motion, and the use of the equation of continuity in a restricted sense serves the needs fairly well. However, the more general hydrodynamical equations are serviceable in a complete study of the motion of ground water and of soil moisture.

In a study of capillary phenomena, soils investigators have generally used the term capillary attraction in a qualitative sense only. The measurement of this attraction in soils is not readily accomplished by direct methods. However, hydrodynamics makes it possible to measure resultant capillary attraction by indirect means, as is shown later. It is generally known that pressure differences from point to point in a liquid give rise to motion from points of high to those of low pressure where other forces such as gravity are not involved. For example, consider the flow of water in a level pipe line. Here the driving force is dependent upon the space rate of change of pressure. The influence of pressure differences on the flow of liquids has an important bearing on the study of the movement of capillary moisture.

In addition to a consideration of the fundamental hydrodynamical equations, the primary purpose of this paper is briefly to review the

efforts which have been made to apply the principles of hydrodynamics to the solution of soil-moisture problems and to indicate the possible outlook for future investigations. The preliminary analysis presented herewith is somewhat general in character and applies both to gases and liquids. The application of the more general equations to irrigation and drainage problems permits some restrictions which are pointed out as the work proceeds. Before developing the general equation of motion, and the equation of continuity in its general form, brief reference is made to the forces which influence the flow of water in open channels and in pipes.

Among irrigation and drainage engineers, it is common knowledge that the velocity of water flowing in open channels and in pipes is determined by two classes of forces, namely, the driving forces and the resisting forces. When a headgate is suddenly opened, permitting water to enter a canal, its velocity is accelerated because the driving forces, F_d, in a down-stream direction parallel to the canal bed at the outset are greater than the resisting forces, F_r, in the opposite direction. The resisting forces are dependent on the velocity since without motion, there is no friction and F_r is zero. For the velocities encountered in most open channels, F_r seems to vary directly with the square of the velocity, and, as the velocity increases, F_r is increased until finally it becomes equal to F_d. According to Newton's Second Law, letting "a" equal the acceleration in the direction of flow we may write for a given elemental volume of water of mass "m," the equation

$$ma = F_d - F_r \quad \text{.....(1)}$$

It is evident from equation (1) that when F_r equals F_d, "a" equals zero. Using this basic equation, together with the fact that the resisting forces are a function of the velocity, Ganguillet and Kutter developed the well-known Chezy-Kutter formula, $V = C\sqrt{RS}$ in which,

V = velocity, usually designated in feet per second.
C = the Kutter coefficient which is a function of the roughness of the channel "n," and also of R and S.
R = the hydraulic mean radius in feet.
S = the slope.

Further attention is given this equation after a consideration of the basic elements of hydrodynamics. The fundamental hydrodynamical analysis which follows is not new to science but applications of the hydrodynamical equations and methods to irrigation and drainage problems are essentially new.

FUNDAMENTAL HYDRODYNAMICAL ANALYSIS

Consider the motion of an elemental volume of fluid in a frictionless medium under the influence of extraneous forces, such as gravity, and also of resultant pressure forces.

The acceleration of a moving particle at a point P, the coördinates of which are x, y, z, is found as follows: At the point P the particle has a velocity V and component velocities of V_x, V_y, and V_z. From P the particle moves to Q, the coördinates of which are $x+\delta x$, $y+\delta y$, $z+\delta z$, in the time δt. In going from P to Q the particle is "taking a step forward in space and also a step forward in time." The increase in the X component, V_x, of the velocity, V, is therefore the sum of the products obtained by multiplying the rates of increase of V_x in the x, the y, and the z directions by the displacements in the respective directions, plus the rate of increase with time multiplied by the interval of time. In mathematical language this change in the X component of the velocity is

$$\delta V_x=\frac{\partial V_x}{\partial x}\delta x+\frac{\partial V_x}{\partial y}\delta y+\frac{\partial V_x}{\partial z}\delta z+\frac{\partial V_x}{\partial t}\delta t \qquad \text{.....(2)}$$

As the change in velocity δV_x occurred in the interval of time δt, dividing equation (2) by δt gives the time rate of change of velocity, which is the acceleration.

Therefore, the X component of the acceleration is

$$a_x=\frac{\delta V_x}{\delta t}=\frac{\partial V_x}{\partial x}\frac{\delta x}{\delta t}+\frac{\partial V_x}{\partial y}\frac{\delta y}{\delta t}+\frac{\partial V_x}{\partial z}\frac{\delta z}{\delta t}+\frac{\partial V_x}{\partial t} \qquad \text{.....(3)}$$

But since by definition

$$V_x=\frac{\delta x}{\delta t},\quad V_y=\frac{\delta y}{\delta t},\text{ and }\quad V_z=\frac{\delta z}{\delta t}$$

equation (3) becomes, after substituting values of $\delta x/\delta t$, $\delta y/\delta t$ and $\delta z/\delta t$

$$a_x=V_x\frac{\partial V_x}{\partial x}+V_y\frac{\partial V_x}{\partial y}+V_z\frac{\partial V_x}{\partial z}+\frac{\partial V_x}{\partial t} \qquad \text{.....(4)}$$

Consider now the pressure forces acting on the elemental volume in a moving fluid. The resultant pressure on the elemental volume in the X direction may be found by reference to a rectangular parallelopiped within the fluid as illustrated in figure 1.

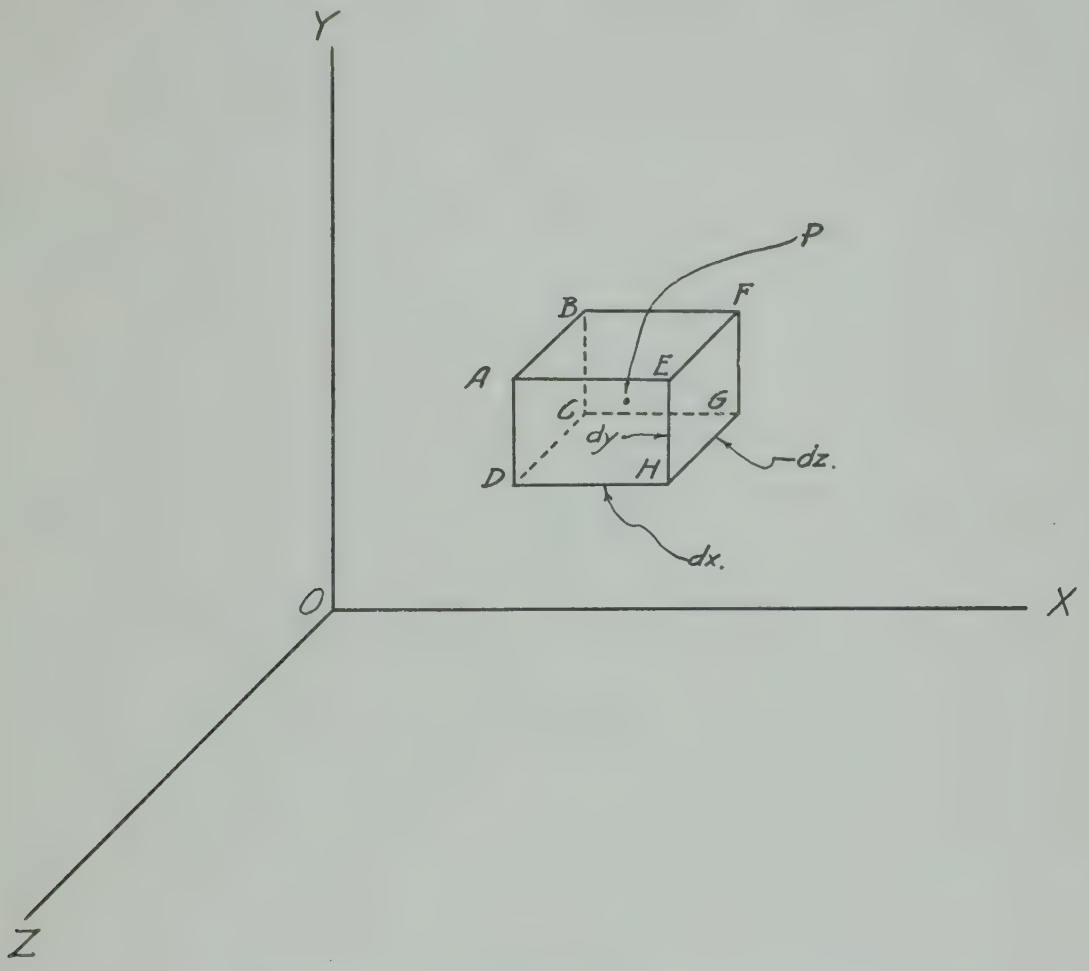

Fig. 1. An elemental volume within a moving fluid.

Let p be the pressure at the center of the elemental volume of which the lengths of the edges are dx, dy, dz. Let ρ be the density, or the mass in unit volume, and let V_x, V_y, V_z, be the component velocities at the point P. The average intensity of pressure on the face $ABCD$ is $\left(p-\frac{\partial p}{\partial x}\frac{dx}{2}\right)$, hence, the total pressure on the face $ABCD$ is $\left(p-\frac{\partial p}{\partial x}\frac{dx}{2}\right)dydz$ .....(a). Likewise the total pressure on the face $EFGH$ is $-\left(p+\frac{\partial p}{\partial x}\frac{dx}{2}\right)dydz$ .....(b); i.e., directed toward the yz plane. Hence, the resultant pressure in the x direction, found by adding (a) and (b), is $-\frac{\partial p}{\partial x}dxdydz$. Let F_x, F_y, and F_z be the components of the extraneous forces per unit mass of the fluid. The mass in the elemental volume is $\rho dxdydz$ and hence $F_x\rho dxdydz$, $F_y\rho dxdydz$, and $F_z\rho dxdydz$ are the resultant components of the extraneous forces on the elemental volume. Now as dx, dy, and dz may arbitrarily be

made very small, the elemental volume may be considered as a particle and hence by Newton's Second Law there results for the X direction,

$$\rho dxdydz\, a_x = \left(\rho F_x - \frac{\partial p}{\partial x}\right) dxdydz \quad \text{.....(5)}$$

The right hand member of (5) is the sum of the resultant X components of the forces on the elemental volume. Dividing by the elemental mass and substituting from (4) the value of a_x gives

$$\frac{\partial V_x}{\partial t} + V_x\frac{\partial V_x}{\partial x} + V_y\frac{\partial V_x}{\partial y} + V_z\frac{\partial V_x}{\partial z} = F_x - \frac{1}{\rho}\frac{\partial p}{\partial x} \quad \text{.....(6)}$$

Equation (6) is the general equation of motion in the x direction for fluids in which friction is neglected. It is apparent that equations for motion in the y and z directions may be similarly derived.

In the analysis of the motion of perfect fluids two classes of motion are encountered, namely, rotational and irrotational motion. For the purpose of this paper only irrotational motion need be considered, and in this class of motion the velocity at any point may be derived from a potential. It is, therefore, desirable to define the term potential, and to consider somewhat fully the meaning of potentials, before applying the general equation of motion to irrigation problems.

The potential at any point P is defined as the negative line integral of the vector from some reference point to the point P, provided the magnitude of this integral has only one value. In mathematical language the potential is, therefore,

$$\Phi = -\int_{P_0}^{P} (E_x dx + E_y dy + E_z dz) \quad \text{.....(7)}$$

where Ex, Ey, and Ez are the components of the vector E along the X, Y, and Z axes respectively.

It follows from the definition of a potential that in a gravitational field every point is characterized by a gravitational potential, in an electrical field by an electrical potential, in a capillary field or a moist soil by a capillary potential. The above potentials, together with some others, may be classed as a group of energy potentials since the line integrals are summations of force multiplied by distance. Under certain conditions, as briefly mentioned above, the velocity at every point is derivable also from a potential. The potential from which the velocity may be derived is called a "velocity potential." It is subject to all the mathematical operations of the energy potentials but differs in some properties as will be shown later.

The energy potentials as commonly used are further defined as the work done *on* existing forces in bringing unit mass from a specified reference point to any other point. For example, consider the work done on gravitational forces in moving unit mass from one point to another under the following conditions. Select a system of two bodies, one having a mass M and one having unit mass. When separated by a distance r, the attraction between these two bodies according to the inverse square law is $E = -\frac{kM}{r^2}$ where k is a constant, depending for its magnitude on the units used and considering r positive from M toward the unit mass. Therefore the work done *on* the gravitational forces in bringing the unit mass a differential distance dr directly toward the mass M is $dw = -\left[-\frac{kM}{r^2}dr\right]$.

The work W done on the gravitational forces in bringing the unit mass directly from the point P_0 to the point P distant respectively R_0 and R from M, therefore, is

$$W = -\int_{R_0}^{R} \frac{kM}{r^2}dr = -kM\left[\frac{1}{R} - \frac{1}{R_0}\right] \quad \text{.....(8)}$$

Let the point P_0 be an infinite distance from M and equation (8) becomes

$$W' = -kM\left[\frac{1}{R} - \frac{1}{\infty}\right] = -\frac{kM}{R} \quad \text{.....(8a)}$$

In the gravitational region infinity has been selected as the reference point. Equation (8a), therefore, gives the gravitational potential at the point P, which is distant R from M, i.e., the work done on the gravitational forces in bringing unit mass from *infinity* to the point in question. A clear conception of the gravitational potential, which is determined by the familiar force of attraction, the magnitude of which is expressed by the inverse square law, helps one understand the meaning of the capillary potential.

The capillary potential at any point P, as here used, is defined as the work done *on* the capillary attraction in bringing *unit mass* of water from the level water surface to any point P within the capillary region. For example, select a column of soil, the water content of which is in equilibrium with the gravitational water in the reservoir, as illustrated in figure 2.

The system being in static equilibrium there is no movement of water in the soil column. Therefore, if we neglect friction, a very

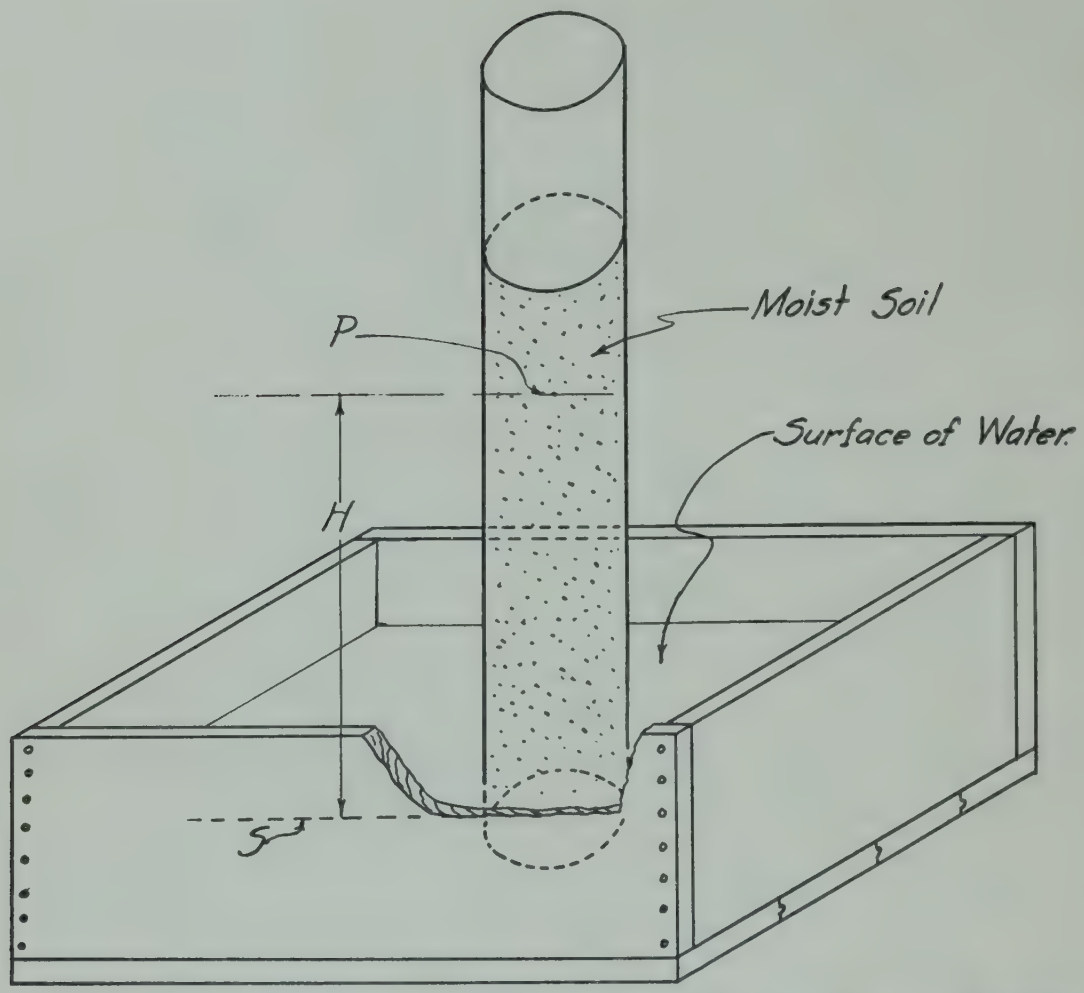

Fig. 2. A column of soil with the water content in equilibrium with the gravitational water in the reservoir.

slight upward impulse applied to unit mass of water at the level of the surface, S, will cause it to rise without having had work done on it to any point P distant H above S. The magnitude of the resultant capillary force as a function of H in bringing the unit mass from the water surface S to the point P is not known, whereas the gravitational force as a function of r in bringing unit mass from ∞ to the point P is known. But as the capillary water is in equilibrium under the action of both the gravitational and the capillary forces it is known that any gain in one form of energy by the unit mass must be equal in magnitude to a simultaneous loss in the other form of energy. The work done *on* the gravity forces, since the integration is in the positive H direction and against the force of gravity, is

$$W_g = -\int_o^H -g\,dH = gH^* \qquad \text{.....(9)}$$

* As H is small the variation in g with H is negligible.

The system being in equilibrium, W_g must be equal in magnitude and opposite in sign to the work ψ done on the capillary forces, because the sum of the two quantities of work must be zero. Therefore,

$$W_g+\Psi=0 \quad \text{or} \quad -\Psi=W_g=gH \qquad \text{.....(9a)}$$

Equation (9a) makes it possible to compute the capillary potential Ψ at every point in a moist soil in which the capillary water is known to be in equilibrium with gravitational water. The capillary potential is a magnitude which characterizes every point in the moist soil.

In the light of the above consideration of potentials, it is desirable to see how the general equation of motion is related to potentials.

The Equation of Motion and Potentials: The vector components E_x, E_y, and E_z of equation (7) may represent component velocities, and hence, where Φ = a velocity potential,

$$\Phi=-\int(V_x dx+V_y dy+V_z dz) \qquad \text{.....(10)}$$

Partial derivatives of Φ with respect to x, y, and z, give the equations:

$$\left.\begin{aligned} \frac{\partial\Phi}{\partial x} &= -V_x \quad \text{.....(a)} \\ \frac{\partial\Phi}{\partial y} &= -V_y \quad \text{.....(b)} \\ \frac{\partial\Phi}{\partial z} &= -V_z \quad \text{.....(c)} \end{aligned}\right\} \qquad \text{.....(11)}$$

Knowing that the order of differentiation is immaterial in successive partial derivatives, there results:

$$\left.\begin{aligned} \frac{\partial}{\partial y}\left(-\frac{\partial\Phi}{\partial x}\right)&=\frac{\partial}{\partial x}\left(-\frac{\partial\Phi}{\partial y}\right) \quad \text{.....(a)} \\ \frac{\partial}{\partial z}\left(-\frac{\partial\Phi}{\partial y}\right)&=\frac{\partial}{\partial y}\left(-\frac{\partial\Phi}{\partial z}\right) \quad \text{.....(b)} \\ \frac{\partial}{\partial x}\left(-\frac{\partial\Phi}{\partial z}\right)&=\frac{\partial}{\partial z}\left(-\frac{\partial\Phi}{\partial x}\right) \quad \text{.....(c)} \end{aligned}\right\} \qquad \text{.....(12)}$$

Substituting from equations (11) into equations (12), it is evident that:

$$\left.\begin{aligned} \frac{\partial}{\partial y}V_x&=\frac{\partial}{\partial x}V_y \quad \text{.....(a)} \\ \frac{\partial}{\partial z}V_y&=\frac{\partial}{\partial y}V_z \quad \text{.....(b)} \\ \frac{\partial}{\partial x}V_z&=\frac{\partial}{\partial z}V_x \quad \text{.....(c)} \end{aligned}\right\} \qquad \text{.....(12a)}$$

Substituting from equation (12a), the values of $\frac{\partial V_x}{\partial y}$, $\frac{\partial V_x}{\partial z}$ in the general equation of motion (6) and also the value of $V_x = -\frac{\partial \Phi}{\partial x}$ from (11) there results

$$V_x\frac{\partial V_x}{\partial x}+V_y\frac{\partial V_y}{\partial x}+V_z\frac{\partial V_z}{\partial x}-\frac{\partial}{\partial t}\left(\frac{\partial \Phi}{\partial x}\right)=F_x-\frac{1}{\rho}\frac{\partial p}{\partial x} \quad \ldots\ldots\ldots\ldots (13)$$

The negative term on the left of the equality sign in (13) may be expressed in the form $-\frac{\partial}{\partial x}\left(\frac{\partial \Phi}{\partial t}\right)$. Further, assuming that F_x may be derived from a potential, it is clear that $F_x = -\frac{\partial \Omega}{\partial x}$ where Ω is a potential due to extraneous forces characteristic of every point in the region. Substituting the above values in (13) shows that

$$\frac{\partial}{\partial x}\left(\frac{1}{2}V_x^2+\frac{1}{2}V_y^2+\frac{1}{2}V_z^2\right)-\frac{\partial}{\partial x}\left(\frac{\partial \Phi}{\partial t}\right)=-\frac{\partial \Omega}{\partial x}-\frac{1}{\rho}\frac{\partial p}{\partial x} \quad \ldots\ldots\ldots\ldots (13a)$$

Letting $V =$ the resultant velocity in (13a) or $V^2 = V_x^2+V_y^2+V_z^2$ multiplying by ∂x and integrating with respect to space keeping time constant there results

$$\frac{1}{2}V^2-\frac{\partial \Phi}{\partial t} = -\Omega-\int\frac{\partial p}{\rho}+C \quad \ldots\ldots\ldots\ldots (14)$$

But the constant of integration C is an arbitrary function of time, i.e., $C = f(t)$. Since precise determination of the velocity potential Φ can be made only with reference to a particular time, t, because the motion may not be steady, it is consistent to consider the C in (14) included in the $\frac{\partial \Phi}{\partial t}$ term and hence from (14),

$$\int\frac{\partial p}{\rho}=\frac{\partial \Phi}{\partial t}-\frac{1}{2}V^2-\Omega \quad \ldots\ldots\ldots\ldots (15)$$

Equation (15) rests on Newton's fundamental law of motion with but two assumptions:

(1) That the extraneous force F_x may be derived from a potential.
(2) That the motion is irrotational.

Two further assumptions are now introduced, namely:

(3) That the density is constant as in water, and
(4) That the motion is steady or does not change with time.

Under assumption (3) $\int\frac{\partial p}{\rho}=\frac{1}{\rho}\int \partial p = p/\rho + a$ constant.

Under assumption (4) $\frac{\partial \Phi}{\partial t} = 0$, since Φ at a particular point is constant with respect to time. Introducing these conditions in (15) there results

$$\Omega + \frac{p}{\rho} + \frac{1}{2} V^2 = \text{constant} \quad \ldots\ldots\ldots\

of flow across this face in unit time is $\left[\rho V_x+\frac{\partial}{\partial x}(\rho V_x)\frac{dx}{2}\right]dydz$. Subtracting the latter mass from the former, the rate of gain in mass between the faces *ABCD* and *EFGH* is found to be equal to

$$-\frac{\partial}{\partial x}(\rho V_x)\,dxdydz \qquad \text{.....(a)}$$

Comparing similarly the flow across the face *CDHG* and *BAEF* it is found that the rate of gain is equal to

$$-\frac{\partial}{\partial y}(\rho V_y)\,dxdydz \qquad \text{.....(b)}$$

For the faces *CBFG* and *DAEH* the rate of gain is equal to

$$-\frac{\partial}{\partial z}(\rho V_z)\,dxdydz \qquad \text{.....(c)}$$

Adding equations (a), (b), and (c) gives the rate of change of mass in the elemental volume. Since mass can be neither created nor destroyed it is known that the sum of (a), (b), and (c) must be equal to the time rate of change of mass within the volume and hence it follows that

$$\frac{\partial}{\partial t}(\rho dxdydz)=-\left[\frac{\partial}{\partial x}(\rho V_x)+\frac{\partial}{\partial y}(\rho V_y)+\frac{\partial}{\partial z}(\rho V_z)\right]dxdydz \qquad \text{.....(d)}$$

Dividing (d) by *dxdydz* and transposing there results the equation of continuity in general form, i.e.,

$$\frac{\partial \rho}{\partial t}+\frac{\partial}{\partial x}(\rho V_x)+\frac{\partial}{\partial y}(\rho V_y)+\frac{\partial}{\partial z}(\rho V_z)=0 \qquad \text{.....(19)}$$

APPLICATION TO OPEN CHANNELS, PIPES, AND SOIL MOISTURE

The movement of moisture in soils is influenced by forces analogous to those which control the flow of water in open channels and pipes. The following brief analysis of well-known formulae in hydraulics, together with some analogies to a proposed soil moisture formula may be helpful in the study of moisture flow.

Open Channels.—The Chezy-Kutter formula $V=C\sqrt{RS}$ heretofore briefly mentioned, was derived from the general equation of motion for the condition of steady flow in which the driving forces are equal and opposite in direction to the retarding forces. Moreover, as indicated below, this formula rests on the fact that the velocity is determined by:

(1) the conductivity, which is dependent upon the form of the channel, and the roughness of surface, and,

(2) The rate of change of energy per unit mass in the direction of flow, or the component of the potential gradient parallel to the surface of the stream.

Consider a stream channel of uniform cross-section, the bottom of which makes an angle α with the horizontal (or sea level), as illustrated in figure 3. Select any length of canal l, from P_0 in the horizontal surface to any point P distant h above the horizontal. Obviously $h=l \sin \alpha$, and therefore from (9) the gravitational* potential φ at P is:

$$W_g=\varphi=-[-gl \sin \alpha] \quad \text{.....(20)}$$

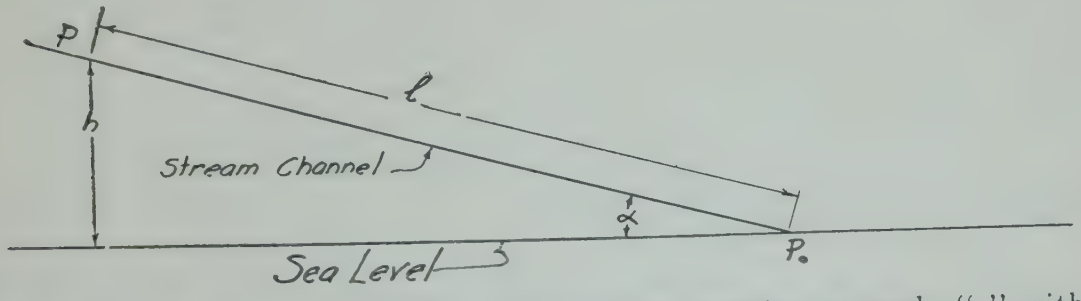

Fig. 3. Stream channel of uniform cross-section, making an angle "α" with a horizontal line.

The gravitational acceleration constant g is implicit in the Chezy-Kutter coefficient C and therefore $C=C_1\sqrt{g}$ where C_1 is a constant which is determined largely by the roughness of the channel. The velocity of water, according to the Chezy-Kutter formula, is

* For the gravitational potential as used in the following analysis, a point in the surface of a body of water or water table is taken as the reference instead of ∞ as in equation (8a). The integration is now opposite in direction to the force of gravity and the potential becomes positive.

$$V = C_1 \sqrt{R}\ \sqrt{gS} = C_1 \sqrt{R}\ \sqrt{\frac{gh}{l}} \qquad \text{.....(21)}$$

The component of the potential gradient in the direction of the velocity is

$$\frac{\partial \varphi}{\partial l} = \frac{\partial}{\partial l}(gl \sin \alpha) = g \sin \alpha = g\frac{h}{l} \qquad \text{.....(22)}$$

Therefore, from (21) and (22)

$$V = C_1 \sqrt{R}\ \sqrt{\frac{\partial \varphi}{\partial l}} \qquad \text{.....(23)}$$

The term C_1 of (23) involves the roughness of channel, the hydraulic mean radius R is the ratio of cross section area to the wetted perimeter and thus involves the form of the channel, and the expression $C_1\sqrt{R}$ is the conductivity, whereas the $\frac{\partial \varphi}{\partial l}$ is the component of the potential gradient along l.

Pipes.—The Weisbach formula commonly used for flow of water in pipes, in a similar manner, may be shown to consist of a conductivity factor and the component of a potential gradient. This formula is:

$$h_f = f\frac{l}{d}\ \frac{V^2}{2g} \qquad \text{.....(24)}$$

where:

h_f = loss of head in friction measured in feet.
f = a friction factor dependent on roughness of pipe.
l = length of pipe in feet.
d = diameter of pipe in feet.
V = the velocity in feet per second.
g = the gravitational acceleration constant.

Solving (24) for V^2 there results:

$$V^2 = \left(\frac{2d}{f}\right)\left(\frac{gh_f}{l}\right) \qquad \text{.....(25)}$$

and for pipes running full the hydraulic mean radius $R = \frac{A}{P} = \frac{d}{4}$ and hence from (25) for flow under pressure,

$$V = 2\sqrt{\frac{2R}{f}}\ \sqrt{\frac{gh_f}{l}} \qquad \text{.....(26)}$$

The factor $2\sqrt{\frac{2R}{f}}$ of (26) involves the form of pipe and its roughness and is the conductivity. The symbol h_f of (26) is analogous to the h of (21) and hence the factor $\frac{gh_f}{l}$ of (26) is the component of the potential gradient.

The above brief analysis concerning the Chezy-Kutter and the Weisbach formulae, it is hoped, will help make clear to irrigation engineers and investigators the purpose of, and the promise in, the application of hydrodynamics to other irrigation problems such as the movement of soil moisture.

Soil Moisture.—It is apparent by analogy from the foregoing illustration that soil moisture flow also is largely determined by two factors, namely:

(1) The conductivity of the soil, and

(2) The potential gradient within the moisture region.

As there are no restrictions on the direction of moisture movement, the flow will occur in the direction of the maximum rate of change of potential. However, the capillary stream is influenced by two potentials, namely: the capillary potential, Ψ, due to capillary pressure; and the gravitational potential, φ, due to gravity. We have, therefore, as a general equation for velocity of the capillary stream,

$$V = C[\nabla^*(\Psi + \varphi)]^m \quad \text{.....(27)}$$

in which the conductivity factor is C and the potential gradient factor is

$$[\nabla(\Psi + \varphi)]^m.$$

For the capillary stream the exponent m is considered equal to unity for reasons given in the following section. The gradient of the potential due to gravity $\nabla\varphi$ being known, it is necessary only to measure Ψ at a few points, from which the component of $\nabla\Psi$ in the direction considered, may be determined. This possibility is further outlined after presenting a description of the method of measuring the capillary potential and a discussion of its relation to the soil moisture content.

There remains then the difficult but not insurmountable task of evaluating the conductivity, C, in (27) for typical soils under particular conditions of soil compactness, temperature, composition of soil water, moisture content and so on.

With the conductivity known, the measurement of the capillary potential, together with its relation to the moisture content would make possible the intelligent attack of irrigation and drainage problems.

Some of the more important of these problems, together with the methods of attack, are considered later. In the next few pages, the progress which has been made in the application of hydrodynamics to ground water and soil moisture is briefly reviewed.

* The symbol ∇ as here used is the gradient, i.e., the rate of change in the direction of the greatest rate of change.

GROUND WATER AND SOIL MOISTURE MOVEMENT INVESTIGATIONS BASED ON HYDRODYNAMICS

Ground Water.—The fruitfulness of dynamics in the study of the flow of heat, electricity, and perfect fluids has long been recognized. The more recent application of dynamics to the motion of ground water was made by Slichter[17] in 1897. Slichter's analysis is based on the equation of continuity (19) and on experimental observations of Darcy, Poiseuille,* and others. These investigators found, as quoted by Slichter,[17] that "the velocity of the flow of a liquid in a given direction through a column of soil is directly proportional to the difference in pressure at the ends of the column and inversely proportional to the length of the column." Mathematically expressed this law is

$$V = k\nabla p \quad \text{.....(28)}$$

in which

v = the velocity
p = the pressure
∇p = the gradient of p, or the rate of change of p in the direction of the greatest rate of change.
k = a constant depending on the size of the soil grains, the soil porosity, the liquid viscosity, and the temperature.

The compressibility of water being very slight, its density ρ may be considered constant, and hence for water $\frac{d\rho}{dt} = 0$ and equation (19) reduces to

$$\frac{\partial V_x}{\partial x} + \frac{\partial V_y}{\partial y} + \frac{\partial V_z}{\partial z} = 0 \quad \text{.....(29)}$$

Using Cartesian coördinates (28) may be expressed as:

$$\left.\begin{aligned} V_x &= k\frac{\partial p}{\partial x} \quad \text{.....(a)} \\ V_y &= k\frac{\partial p}{\partial y} \quad \text{.....(b)} \\ V_z &= k\frac{\partial p}{\partial z} \quad \text{.....(c)} \end{aligned}\right\} \quad \text{.....(30)}$$

* The experimental observation of Poiseuille is confirmed by Lamb[13] in a theoretical analysis of the flow of a liquid through a pipe of uniform section. (Hydrodynamics, 4th ed., § 331, p. 577.)

Substituting the values of V_x, V_y, and V_z from (30) into (29) and dividing by k there results

$$\frac{\partial^2 p}{\partial x^2}+\frac{\partial^2 p}{\partial y^2}+\frac{\partial^2 p}{\partial z^2}=0 \quad \text{.....(31)}$$

Equations (29) and (30) are identical with Slichter's equations (4) and (5) of his chapter II.[17] Equation (31) is known as Laplace's equation and, like the equation of continuity (29) forms, in part, the basis of many important engineering analyses.

The fact that the "velocity potential," which satisfies (31), is proportional to the pressure in ground-water motion was first pointed out by Slichter. Moreover, Slichter made his excellent contribution to the investigation of ground water motion largely by pointing out the coincidence of the pressure function with the velocity potential (omitting the constant k) and by showing that the solution of any problem in the motion of ground water is dependent upon solving the differential equation (31).

Soil Moisture.—The first application of the analytical method to capillary action in soils was made by Buckingham[3] in 1907. He called attention to the fact that the movement of capillary moisture in any soil is dependent on its conductivity and on the driving force. Buckingham also pointed out, after proposing a general equation for measuring the flow of soil moisture, a formal analogy of this equation to the equations for flow of heat as measured by Fourier's law, and for the flow of electricity as measured by Ohm's law. Buckingham's equation is

$$Q=\lambda S \quad \text{.....(32)}$$

in which

Q="The capillary current density at any point, i.e., the mass of water which passes in one second through one square centimeter of an imaginary surface perpendicular to the direction of the flow."

S=The capillary potential gradient ($\nabla\Psi$), or "the amount by which the potential Ψ increases per centimeter in the direction of the current, by reason of the fact that the water content of the soil decreases in that direction."

λ="The capillary conductivity of the soil." The Q in equation (32) is numerically equivalent to the V of (28) hence we may write (32) in the form

$$V=\lambda\nabla\Psi \quad \text{.....(33)},$$

which is analogous to Darcy's experimental law[17] of flow for gravitational water.

Following Buckingham's work but little use was made of the fundamental laws of motion until 1920, when Gardner[7] briefly considered "the capillary potential and its relation to soil moisture constants." Very early in Gardner's soil-moisture studies he called attention to the fact that the capillary potential makes possible a new interpretation of soil-moisture constants, such as the hygroscopic coefficient, the moisture-holding capacity, the saturation constant, and the moisture equivalent. Each of these several constants really define equi-potential regions regardless of variation from one soil to another.

Concerning the definition of capillary potential, in the preliminary paper above referred to, Gardner says "It is perhaps quite immaterial where the zero potential is placed and also what convention is adopted as to the algebraic sign, although it is somewhat more in accord with modern usage to define the potential as the work done by the field forces in bringing unit mass from the point in question to infinity, and in such case the heat of vaporization should be added to Buckingham's potential and the negative sign should be used." However, for measurements of the capillary potential, which have not yet been published, Gardner has selected the water table as zero potential.

Gardner's later analysis of the dynamical problem with special reference to the movement of soil moisture appeared under the joint authorship of Gardner and Widtsoe[8] in 1921. The excellent analysis of these authors is based on the "assumption that the mean velocity of the water through the soil is proportional to the pressure gradient, or more generally, to the force per unit volume." That this assumption is supported both by fundamental analysis and by experiment is pointed out by the authors, as evidenced in part by the following statements:

"For bodies moving in response to conservative forces the resultant of the effective [external] forces is a measure of their acceleration in the direction of the resultant.* Where friction comes into play, however, this is not true; but a limiting velocity is soon reached when the frictional force becomes equal and opposite to the resultant of the impressed forces. For small velocities of such magnitudes as are encountered in the soil, this frictional force is directly proportional to the velocity. If, for example, water is forced through a small pipe of regular or irregular section, including the pores of a homogenous soil, the mean velocity is found from theory[13] and experiment[17] to vary directly as the pressure gradient† with a proportionality factor which involves the shape and size of the tube."

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* See also equation (1) and accompanying discussion in this paper.

† Some exceptions to this law were noted by F. H. King and recorded in U. S. Geol. Survey 19th Ann. Rept., pt. II. 61-294. *figs. 1-53. pls. VI-XVI.* 1898.

If the observation based on experiment and theory indicating that the frictional forces are directly proportional to the first power of the velocity be accepted as established law in soil moisture flow then equation (34) which is based on the Gardner-Widtsoe assumption may be derived with slight modification from the general equation of motion (6), as will shortly be demonstrated.

The Gardner-Widtsoe[8] equation is

$$V = K\rho\nabla\Phi^* \qquad (34)$$

where

V = mean velocity at a point in the soil.
K = a proportionality constant.
ρ = moisture density at a point.
Φ = the sum of three potentials, π, Ψ, and φ; where
π = potential due to hydrostatic pressure.
Ψ = potential due to capillary pressure.
φ = potential due to gravity.
∇ is a mathematical operator written thus

$$\nabla \equiv i\frac{\partial}{\partial x} + j\frac{\partial}{\partial y} + k\frac{\partial}{\partial z}$$

Referring again to the general equation of motion in frictionless media (6), it is apparent that for a steady state, or for acceleration so small as to be negligible, the left-hand member is zero. Applying the general equation (6) to the flow of soil moisture in any direction and in which there is friction and letting F_r = the resultant of the frictional forces on unit *mass* it is evident from (6) that

$$F - \frac{1}{\rho}\nabla p + F_r = 0 \qquad (35)$$

It is important to note that ρ as used in (35) and the analysis following is considered a variable. Conceive a given finite volume of soil as being composed (1) of a solid-soil phase, (2) a liquid-water phase, and (3) a gas (water-vapor-and-air) phase. If ρ is defined as the mass of phase (2), i.e., the liquid water, per unit volume of the space which is occupied by the three phases, then it is a variable. On the contrary, if ρ is defined as the mass of phase (2) per unit volume of phase (2), it would be substantially constant. The first definition given above, i.e.,

* The negative space rate of change of the energy potentials is work per unit mass divided by length, i.e., $-\nabla\Phi = \frac{\text{Force}}{\rho L^3} \times \frac{L}{L}$ = force per unit mass = F. Therefore, since $-\nabla\Phi = F$ and since there is in unit volume a mass of water, ρ, it follows that the magnitude of the resultant force per unit volume in the direction of V is $\rho F = \rho\nabla\Phi$, or the mass in unit *volume* times the force per unit *mass*.

the mass of liquid water per unit volume of space, is the one here used, and the applicability of the hydrodynamical equation of motion to the flow of soil moisture rests in part on the correctness of the concept of ρ as a variable. It rests also on the belief that the apparent discontinuities in the liquid-water phase may be ignored since in reality every particle of soil, or every small group of particles, is completely surrounded by capillary water and in this sense the liquid-water is continuous. It follows from the definition of ρ here used that V must signify the mean velocity of the liquid-water particles.

The quantity (mass) of liquid water, Q, that flows in unit time across a unit surface within the soil and normal to V, is given by the equation

$$Q=\rho V^* \quad \text{.....(36)}$$

According to the above definition of ρ, it is evident that as ρ increases the capillary pressure decreases and therefore the capillary pressure is a function of the density ρ.

The only extraneous force influencing the moisture flow is gravity, and from the preceding discussion of potentials $F=-\nabla\varphi$ where φ is the potential due to gravity. Also, since the resisting forces per unit *volume* are directly proportional to the first power of the velocity, $\rho F_r=aV$ where "a" is a constant characteristic of the soil for a particular moisture content. Substituting these values of F and F_r in (35) there results

$$\rho\nabla\varphi+\nabla p=aV \quad \text{.....(37)}$$

In the general case of soil water movement there are two kinds of pressure, namely:

p_c = capillary pressure or the negative pressure or tension due to curved surfaces, and

p_h = hydrostatic pressure.

The pressure p in (37) is equal to the sum of p_c+p_h . . . . hence

$$aV=\nabla(p_c+p_h)+\rho\nabla\varphi \quad \text{.....(38)}$$

Since $\nabla\Phi$ is force per unit *mass* and ρ = mass per unit volume, it follows that $\rho\nabla\Phi$ is force per unit *volume* and that

$$\rho\nabla\Phi=\nabla(p_c+p_h)+\rho\nabla\varphi \quad \text{.....(39)}$$

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* If, however, ρ is defined as the mass of phase (2) per unit volume of phase (2), then its magnitude would be unity (approximately) with the C. G. S. units. Under this condition the quantity (mass) of liquid water, Q, that flows in unit time across a unit surface within the soil and normal to V, is given *numerically* by the equation:

$$Q=rV$$

Where V is the mean velocity of the liquid water particles as above defined and r is the cross-section area of the liquid-water capillaries in unit surface within the soil and normal to these capillaries.

Hence (38) may be written by substituting from (39)

$$V = k\rho\nabla\Phi \quad \text{.....(40)}$$

in which $k = \frac{1}{a}$. Equation (40) being the same as the Gardner-Widtsoe equation (34) indicates that the latter equation is in accord with the general equation of motion.

It is important to observe that Slichter's basic equation (28) for the flow of "gravitational" ground water and the Gardner-Widtsoe basic equation for the movement of soil moisture (34) are formally the same. The ∇p factor in (28) is equivalent to the $\rho\nabla\Phi$ factor in (34). Moreover, as shown by Gardner and Widtsoe, under the condition of complete saturation of the soil, at which time the capillary pressure is zero and the time rate of change of the mass per unit volume ρ is zero, their general equation of motion for capillary water, i.e., their formula (4) on page 221,[8] reduces to $\nabla^2 P = 0$* which is identical with Slichter's equation (5), page 330, or number (31) in this paper.

Equation (II) of the Gardner-Widtsoe paper, as corrected in the footnote,* is the differential equation derived from the equation of continuity and the equation of motion for a steady state. After the introduction of certain tentative assumptions concerning the value of Ψ in terms of ρ, this equation reduces to

$$\nabla^2\rho + \frac{2\rho}{c}\nabla\varphi\nabla\rho = 0 \quad \text{.....(41)}$$

which "is the most common condition met with in irrigation practice." Equation (41), together with others deduced from the general equation, including that for the condition of horizontal capillary flow under the influence of capillary forces only, and others for other conditions, are shown by Gardner to be in fairly close agreement with experimental data.

There is, however, in the movement of soil moisture an additional factor the influence of which, as yet, is not evaluated in the equations above reviewed, which may exert a significant influence, namely, the concentration of moisture or degree of saturation. Gardner and

* The Gardner-Widtsoe analysis on page 221[8] might be clarified a little by calling attention to the fact that the mathematical operator ∇ includes the unit vectors i, j, and k and is usually written

$$\nabla \equiv i\frac{\partial}{\partial x} + j\frac{\partial}{\partial y} + k\frac{\partial}{\partial z}.$$

Further, in their case (2) the middle term of the last factor in the right-hand member should be $(\nabla\rho)(\nabla p)$ instead of $(\nabla p)^2$, thus making the corrected equation

$$\frac{\partial\rho}{\partial t} = -K[\rho\nabla^2 p + (\nabla\rho)(\nabla p) + 2\rho\nabla\varphi\nabla\rho]$$

Widtsoe refer to this factor in the following language: "Where the soil is unsaturated and movement takes place in response to capillary forces, it is evident that the degree of saturation may become an additional factor, but in the absence of direct evidence that this has an appreciable effect upon the inherent moisture conductivity (K of equation 34), we may *temporarily* ignore the moisture concentration."

Buckingham[3] also recognized the importance of this factor and devoted considerable space to its consideration.

The design of suitable equipment and the development of methods for measuring the capillary potential has made possible a systematic study of the influence of the moisture concentration ρ on the moisture conductivity.*

MEASUREMENTS OF THE CAPILLARY POTENTIAL

Buckingham[3] measured the capillary potential by making direct readings of the height of rise of moisture in vertical soil tubes. As his method is fully described in available literature it is not further considered here.

For the methods described below I am indebted to Gardner and associates of the Physics Laboratory, Utah Agricultural College Experiment Station. The equipment now used by Gardner, a description of which has not heretofore been published, is shown in figure 4. It consists of a special U-shaped glass tube U; an inverted glass funnel F, to which is cemented a porous porcelain cup P; a bucket of soil S; and a quantity of mercury Hg. The inverted funnel is securely fastened to a connecting tube B, leading to one branch of the U-tube, by means of a tightly fitting vacuum rubber connection R. The connecting tube, B, is similarly fastened to the U tube by a vacuum rubber connection R′. The complete equipment is termed a capillary potentiometer.

The procedure in measuring the capillary potential is substantially as follows: The funnel F, after being cemented to the porous cup P with a mixture of hot asphalt, is attached to a suction pump to ascertain the maximum pressure which the cup will stand without leaking air. The cup is then filled with water, care being exercised to exclude air bubbles, and placed in the soil container as shown in figure 4 and connected at R. The connecting tube B is then filled with water, and the bucket, S, is filled with water up to the elevation of the plane Q, which causes the mercury in the small branch of the U tube to take the position

* An experiment concerning the influence of moisture content on conductivity is being conducted in the laboratory of the Physics Dept., Utah Agr. Exp. Sta.

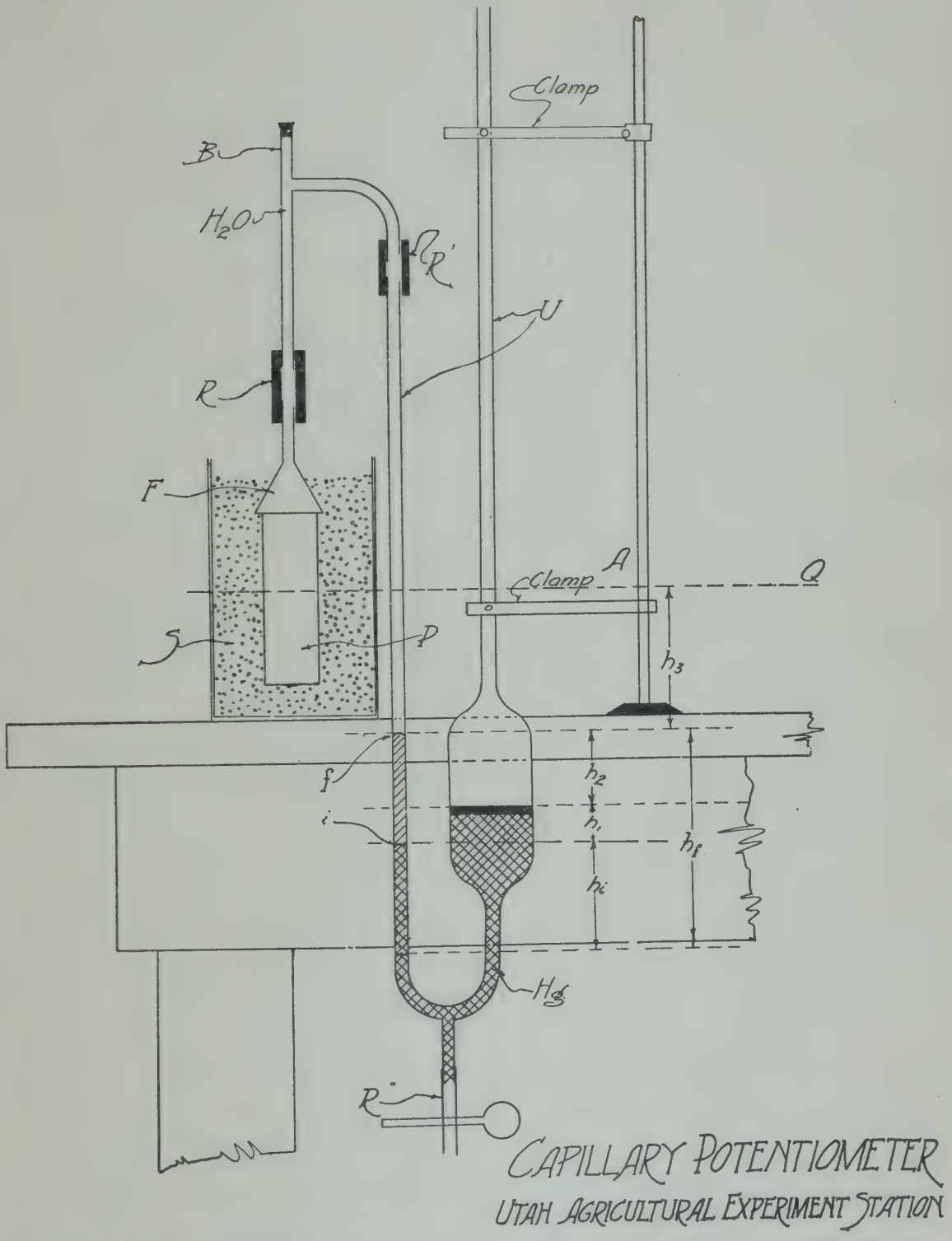

Fig. 4. The capillary potentiometer designed by Gardner, of the Utah Agricultural Experiment Station. This equipment is used in direct determinations. B, connecting tube. F, inverted glass funnel. P, porous porcelain cup. R, R′, R″, vacuum rubber connections. S, bucket of soil. U, U-shaped glass tube.

i. The water is then taken out of the bucket, and soil of the desired moisture content* is packed around the cup. The water is drawn by the soil through the walls of the porous cup. Thus the capillary forces do work against the force of gravity on the mercury by changing the mercury levels in the U tube from the initial position i, i.e., the position at which the mercury would rest if the porous cup were surrounded by water up to the plane Q, to the final position f, which represents the position after the soil has been packed about the cup and equilibrium has been reached.†

The capillary potential, Ψ, when measured in gram centimeters per gram, is numerically equal to the negative hydrostatic pressure measured in grams per square centimeter. Equation (9a) may therefore be written, by employing the above units,

$$-\Psi = H = p_f \quad \text{.....(numerically).....} \quad (42)$$

where p_f = the hydrostatic pressure at a point equivalent to a distance H centimeters above a free water surface after equilibrium has been reached.

At the elevation of the plane Q, the hydrostatic pressure in the U tube of Fig. 4 is atmospheric at the beginning of the capillary potential measurement since the water surface in the soil bucket around the cup is then at the elevation Q. The initial hydrostatic pressure at elevation Q, when the mercury is at i, is therefore equivalent to zero capillary potential. After equilibrium has been reached and the mercury is at f, it is evident from Fig. 4 that

$$p_f + 13.6h_1 = 13.6(h_1 + h_2) + h_3 \quad \text{.....(a)}$$

Remembering that the pressure at elevation Q is atmospheric at the beginning of the measurement, it may be seen from Fig. 4 that when the capillary potential is zero

$$13.6h_1 = (h_1 + h_2) + h_3 \quad \text{.....(b)}$$

Subtracting (b) from (a) there results

$p_f = 12.6(h_1 + h_2)$ and substituting

in (42), it is apparent that

$$\Psi = -12.6(h_1 + h_2) \quad \text{.....(c)}$$

It is therefore necessary with this method only to measure the height of rise of the mercury column and multiply by (-12.6) to obtain the value of Ψ in gram centimeters per gram.

* The capillary potential of Greenville soil having less than 14 per cent moisture has not been measured as yet.

† The cross-section area of the U tube in the enlarged section is 30 times that of the small section, hence the lowering in the large section is ignored.

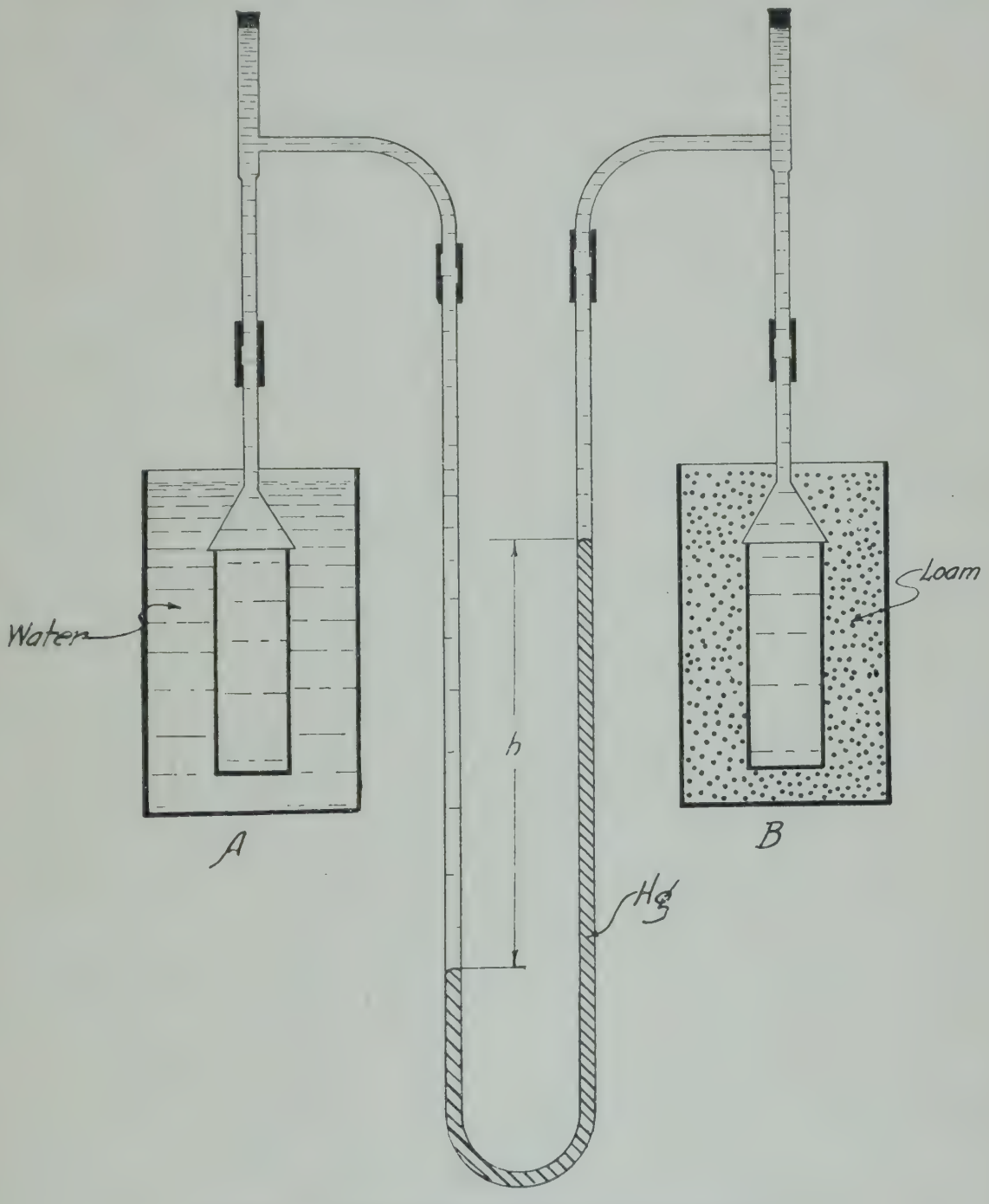

Fig. 5. Differential capillary potentiometer.

In order to measure the potential a differential method also has been developed. The equipment is illustrated in figure 5. It will be noted that bucket A has water outside as well as inside the porous cup. The potential as measured by h is then the absolute value for the soil in bucket B. If soil were placed also in A then h would measure the difference in Ψ between the soil in the two buckets.

Using the differential method just described, 9 buckets A, B, C, D, E, F, G, H, and I were arranged in series as shown in figure 6.

Bucket A was filled with water but contained no soil. The moisture percentage on the oven-dry weight basis ranged from 22.16 in B down

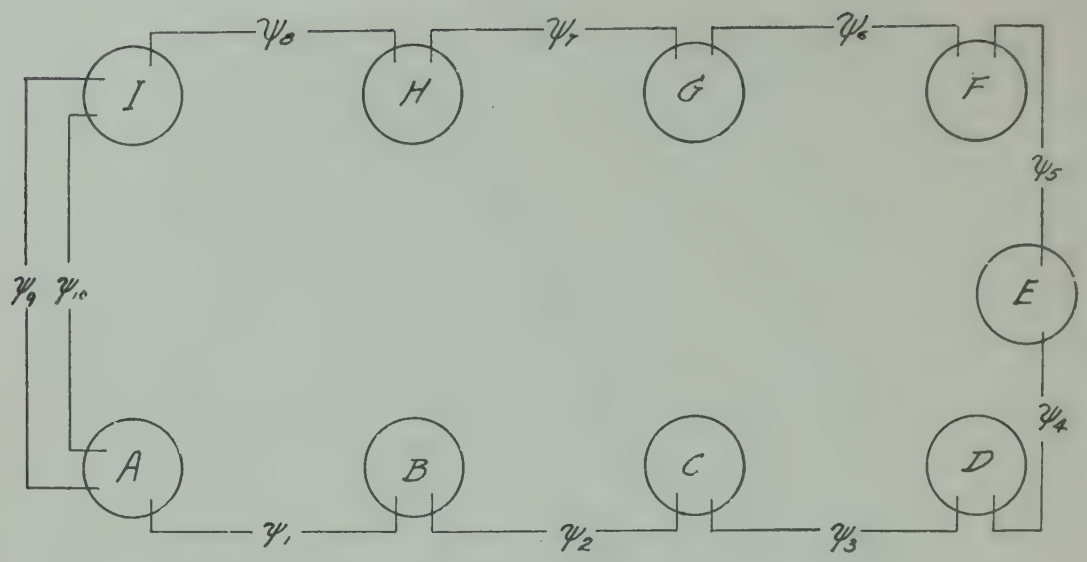

Fig. 6. Typical arrangement of buckets for measuring capillary potential by the differential method.

to 14.58 in Bucket I.* Measurements of the potential difference were made between buckets A and B, B and C, C and D, ... and so on by the potentiometers P_1, P_2, P_3, ... to P_{10}, and recorded as potentials Ψ_1, Ψ_2 . . . Ψ_{10}. The sum of the potential differences Ψ_1 to Ψ_8 should equal Ψ_9 or Ψ_{10} as measured by potentiometer P_9 or P_{10}, the latter two being duplicate measurements. At the end of an eleven-day test, the sum of Ψ_1 to Ψ_8 inclusive was 604 gm.-cm. per gm. and the average of Ψ_9 and Ψ_{10} was 614 gm.-cm. per gm. This is a satisfactory agreement. Further reference is made to the above measurements by the differential method in connection with the following data concerning the influence of the moisture percentage on the capillary potential.

RELATION OF THE CAPILLARY POTENTIAL (Ψ) TO THE MOISTURE CONTENT OF THE SOIL (ρ')†

That the curvature of soil moisture films is influenced by the moisture content of the soil is common knowledge among soils investigators. As the capillary potential Ψ at every point characterizes the film curvature at that point it is clear that the magnitude of Ψ is dependent in part on the moisture content ρ'. Therefore, $\Psi = f(\rho')$ for a given soil compactness, temperature, etc. If the relation $\Psi = f(\rho')$ were established for all conditions it would be possible to write, from (42)

$$V = k\rho\nabla[f(\rho') + \varphi] \quad \ldots \quad (43)$$

* Bucket E contained 24 per cent moisture, but, as one of the cups in this bucket leaked, this moisture content is rejected.

† The term ρ' signifies moisture percentage on the dry weight basis, hence $\rho = A_s\rho'$ where A_s = the apparent specific gravity of the soil.

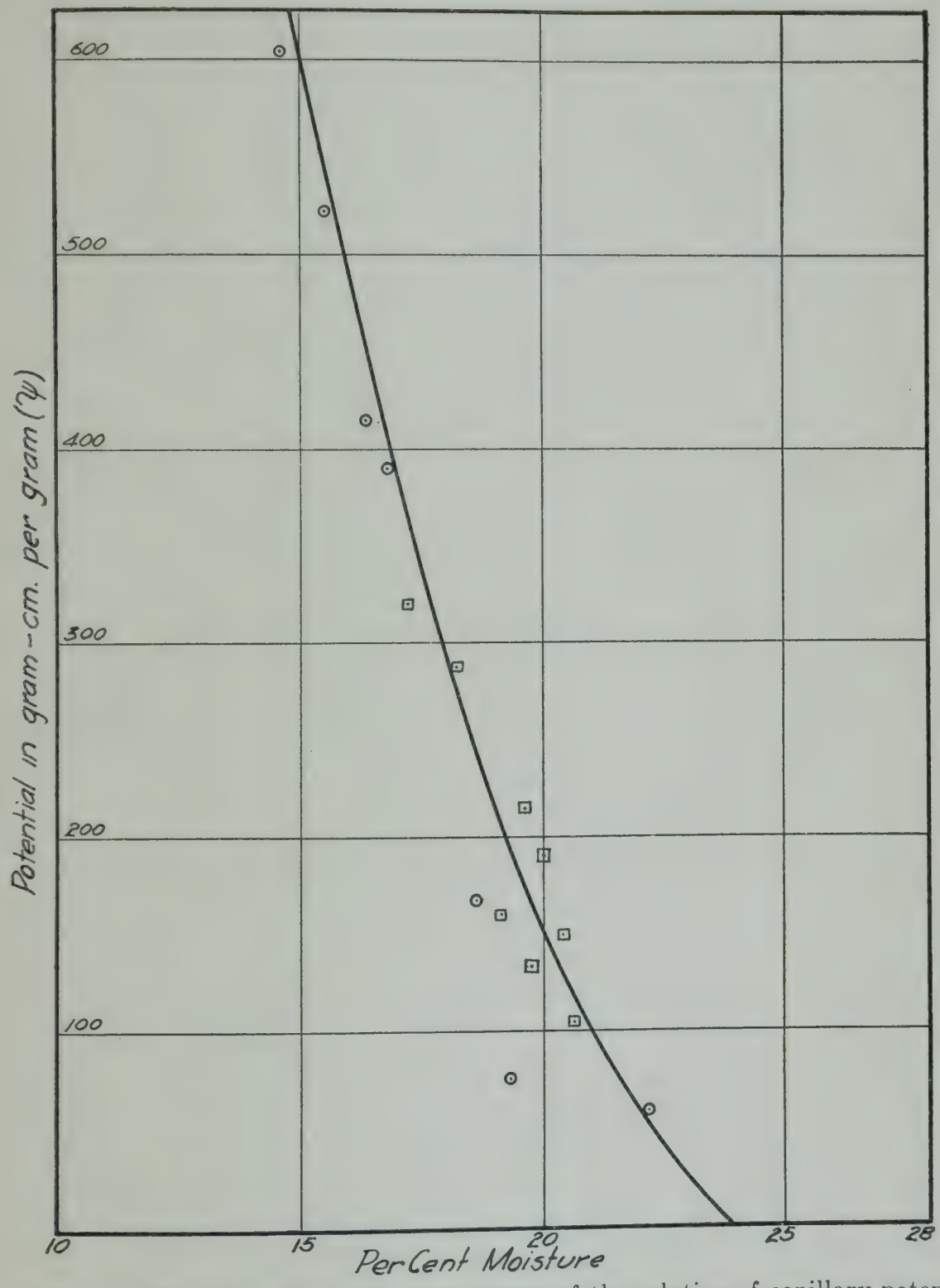

Fig. 7. Typical laboratory measurements of the relation of capillary potential Ψ to the moisture percentage ρ'.

TABLE 1

RELATION OF THE CAPILLARY POTENTIAL Ψ TO THE MOISTURE PERCENTAGE ON THE DRY WEIGHT BASIS ρ' AS INDICATED BY DIRECT METHOD MEASUREMENTS

A	B	C	D	E	F	G	H
Number of bucket	Potentiometer number	Depression of Hg in capillary tube with porous cup in water	Elevation of Hg in capillary tube when porous cup is in soil	Total height of rise of Hg in the capillary tube	Average total height for 3 potentiometers	Capillary potential in gm.-cm. per gram $=12.6(h_1+h_2)$	Moisture percentage based on dry weight of soil
		h_1	$+h_2$	h_1+h_2	$\frac{h_1+h_2}{3}$	Ψ	ρ'
6	a	17.0	8.6	25.6	25.3	319	17.2
	b	15.9	9.1	25.0			
	c	17.3	8.0	25.3			
2	a	17.4	5.8	23.2	22.9	288	18.2
	b	16.7	6.3	23.0			
	c	14.8	7.7	22.5			
7	a	6.2	6.9	13.1	12.7	160	19.1
	b	5.3	6.8	12.1			
	c	6.3	6.6	12.9			
3	a	12.9	4.4	17.3	17.0	214	19.6
	b	11.5	5.4	16.9			
	c	10.6	6.2	16.8			
8	a	2.4	8.5	10.9	10.4	131	19.8
	b	2.3	7.7	10.0			
	c	1.1	9.2	10.3			
1	a	10.2	5.4	15.6	15.4	194	20.0
	b	10.4	5.1	15.5			
	c	7.8	7.4	15.2			
4	a	5.8	6.0	11.8	11.9	150	20.4
	b	4.7	6.9	11.6			
	c	7.5	4.7	12.2			
5	a	2.3	5.9	8.2	8.3	105	20.6
	b	0.9	7.1	8.0			
	c	2.5	6.2	8.7			

From (43) the driving force causing the flow of soil moisture may be determined directly by measuring the space rate of change of ρ' from point to point in the soil.

Some typical laboratory measurements of the relation of Ψ to ρ' are presented in table 1 and also in figure 7. Of the 15 points plotted in figure 7, the eight points inside of the small squares were determined by the direct method of measuring capillary potential first described. The seven points within the small circles were determined by the differential method. Both tests were made with the Greenville soil. To determine the moisture percentage, ρ', all of the soil in each bucket was oven-dried.

Figure 7 indicates that the rate of change of the potential with the moisture content $\frac{d\Psi}{d\rho'}$, is comparatively high for low moisture percentages and low for relatively high moisture percentages. The figure suggests that the potential is high when the moisture content is low and that for large moisture contents, as would be expected, the potential is low.

As the capillary potential is zero at the water table, where the soil is completely saturated, it appears from figure 7 that the potential decreases relatively slowly as the moisture content is increased from 22.2 per cent to the saturation point which is probably well above 25 per cent. A statistical analysis of 84 laboratory determinations of the relation of Ψ to ρ', made by Gardner but not yet published, seems to indicate that over a certain range this relation may be represented by an equilateral hyperbola of the form

$$(\rho'-a)(\Psi+b)=c^* \quad \text{.....(44),}$$

in which a, b, and c are constants that may be evaluated from experimental observations. Moreover, this relation is apparently in accord with natural conditions. It seems, for example, that Ψ may become very large for a moisture content near the wilting point. According to (44) when Ψ is infinite $\rho'=a$ and when ρ' is infinite $\Psi=-b$. That equation (44) precisely represents the (Ψ, ρ) relation is not yet established, and therefore the data presented in table 1 and figure 7 should be interpreted as illustrating the trend of variation of Ψ with ρ'. Further reference is made to the data presented in figure 7 in connection with a study of the field capacity of soils for irrigation water, results of which are presented in figure 14.

* The maximum Ψ as yet measured is slightly over 600 gm.-cm. per gm. and the minimum moisture content is a little over 14 per cent.

SOME APPLICATIONS TO IRRIGATION AND DRAINAGE PROBLEMS

It is not the intent of this paper to enumerate all the ways in which hydromechanics may advantageously be applied to irrigation and drainage problems, nor to evaluate coefficients for the soil moisture velocity equation $V = k\rho\nabla\Phi$. On the other hand, the purpose is to call attention to the fact that use can be made of knowledge concerning equipotential regions and capillary potential measurements in a study of water capacity of soils and of moisture conditions above a high water table.

Irrigation and drainage are simply means of controlling the water content of arable soils for purposes of producing economical crops, the purpose of irrigation being to maintain in the soil an adequate moisture content, and that of drainage to prevent the occurrence of excessive amounts of water. The ideal way to irrigate is to supply water at the same rate as it is needed by plants, but such procedure is impracticable. It is therefore necessary to use the soil as a water reservoir in which there may be stored in a form available to plants the amount of water needed during the time between irrigations. The necessity for such storage, the difficulty in determining the capacity of soils for water, the inherent dread of drought among farmers in an arid climate, the great variation of soil texture and structure, together with other important factors, have led to the application of excessive amounts of irrigation water. Excessive irrigation, seepage from canals, and percolation from high lands to low lands in western valleys have brought about a rise in the water table which has rendered large areas of the best land either partially or wholly unproductive without artificial drainage. During the early years of irrigation, because of the very dry conditions of virgin soil and the great depth to the water table, the ultimate seriousness of wasteful use of water is not apparent. However, after many years of irrigation, when water has become more valuable, when the virgin aridity of the soil has been removed, and the water table has risen to elevations which endanger plant life, it becomes very necessary to have dependable information concerning soil moisture control. The capacity of the soils for water, the movement of soil moisture and of nitrates and other soluble salts, and the relation of drainage to capillary phenomena become increasingly important where irrigation has been practiced many years. That the solution of these problems may be furthered by the application of hydromechanics is pointed out below.

Capacities of Soils for Water.—Many advantages are gained by storing in the soil from a single irrigation all of the water it will retain for plant use, and likewise there are many disadvantages in applying in a single irrigation more water than the soil will hold. Some methods of determining the water capacity of soils have heretofore been reported[10,11] and only those phases of the question not considered previously will be discussed here.

The precision of water capacity measurements rests in large measure on two important factors, which, in general, have not been sufficiently recognized by soil moisture workers, namely:

1. The time after flooding the soil which is selected as representing the moisture capacity, and
2. The depth of the water table, or the reference point for the capillary potential.

The importance of these two factors, in reality, is due to the fact that under the natural conditions encountered in irrigation the soil moisture is seldom at rest. It is very difficult to obtain a condition of equilibrium of moisture in field soils.

It is likewise difficult to ascertain whether or not the moisture in a soil is in a condition of equilibrium. During the period in which equilibrium of moisture conditions is being approached, the velocity of moisture movement is so low that it cannot easily be detected by direct measurement. Since there can be no hydrostatic pressure in unsaturated soils in which capillary pressures exist, it follows that the total potential Φ of equation (40) is equal to the capillary potential, Ψ, plus the gravitational potential, φ. The gradient of the gravitation potential, $\nabla\varphi$, is known to be g. If, therefore, several measurements of the capillary potential, Ψ, at different elevations show that the gradient of the capillary potential $\nabla\Psi = -g$, then the moisture region considered is an equipotential one and that there can be no vertical moisture movement because the driving force is zero. The need of some satisfactory method of detecting the existence of equilibrium of moisture was urgently felt in 1919 in connection with some water capacity studies of soils under natural field conditions at the Utah Experiment Station. The results of these experiments are of interest in showing the difficulty of detecting without the aid of capillary potential measurements at what time the moisture reached a condition of equilibrium. They indicate also that when the condition of equilibrium is approached, after *excessive* irrigation of soils having uniform texture, the moisture content increases as the depth of the soil increases. A non-technical report[11] of these water capacity experiments was published in 1922. A brief statement of the plan of the experiments taken from this report is given below:

"To remove all doubt concerning *completeness of saturation* and also to remove the influence of the growing crop, the authors prepared three rectangular basin plats to which excessive amounts of water were applied. Each plat was 38 feet long and 33 feet wide. Around these plats levees about two feet high were built with soil taken from outside of the plats; thus, the soil in the plats was left undisturbed. The plats were numbered A, B, and C. Samples of soil were taken to ascertain the moisture content before irrigation, after which plat A was given a 12-inch irrigation, plat B a 24-inch irrigation, and plat C a 36-inch irrigation.

"The borings for moisture samples were made to a depth of 12 feet and the moisture determinations were made in the laboratory by the usual methods, the results being recorded in per cent of the weight of the dry soil."

The soil is a deep uniform loam having a water table about 50 feet below the surface. Widtsoe and McLaughlin[19] have published a detailed statement of the chemical and physical properties of the soil.

Observations concerning the movement of capillary water in the three plats, A, B, and C, are presented graphically in figures 8, 9, and 10. The data are reported in acre-inches of water per acre, for different depths of soil. Moisture determinations were made at irregular intervals from June 16 to October 11, 1919; there being 2,556 determinations, of which 1,476 were made in June, 468 in July, and 216 in August and September. The location of borings was systematically made, and stakes were placed in each hole as soon as the sample had been obtained and the excess disturbed soil had been replaced. On each stake the date of sampling was marked, thus avoiding duplication in the location of borings.

On June 16, immediately after the irrigation water disappeared from the surface of the soil, a heavy straw mulch from 8 to 10 inches deep was spread over the surface of each plat. To determine the loss of water through the straw, an evaporimeter pan 12″ by 20″ was filled with soil of about the same moisture content as that in the plat and was placed under the straw in plat A with its surface flush with the ground surface. From August 2 to 26, the pan lost 1,294 grams of water, which is equal to 0.838 cm. depth, or 0.035 cm. a day. Measurement of the decrease in moisture content of the upper 6 feet of soil from June 16 to August 22, after deducting the water evaporated, shows a loss from plats A, B, and C of 0.58, 0.64, and 0.71 cm. respectively, in 24 hours. These measurements are based on six borings in each plat and six samples in each boring.

The averages of the samples for each of the plats on the various dates on which moisture tests were made is presented for three different depths of soil in the curves of figure 8. It will be noted by the rapid decline of the upper curves of figure 8, each point of which is based on 72 moisture determinations, that large amounts of capillary water passed below the 12-foot level in plats B and C shortly after the irrigation. It appears that in plats B and C, and in the upper 6 feet of plat A, the moisture continued to move downward from June 16 to August 22. From June 16 to June 30 in plat A there was a significant increase in the moisture content from 7 to 12 feet in depth, but from July 5 to August 22 there seems to have been a downward movement even from this depth, as the total loss of moisture is greater than the evaporation loss.

The curves for the entire 12-foot depth show that equilibrium was closely approached, if not actually reached, on August 22, because after this date there was practically no further loss. The small gain in October was due to a rainfall of 1.36 inches early in the month.

In figure 9 the loss of moisture for each plat is compared in four depth zones of soil, namely, from 0 to 3 feet, 4 to 6 feet, 7 to 9 feet, and 10 to 12 feet inclusive. The notable features shown by this figure are that in the upper 3 feet of soil, plat A, which received a 1-foot irrigation, was almost as fully wetted at the outset as plat C, which received a 3-foot application. In the second 3 feet of soil the water content in plat A was appreciably less than that in plats B and C, during the first 10 days after irrigation. In the third and fourth 3-foot sections the differences are much greater in magnitude and of longer duration. To illustrate: in the fourth 3-foot section the moisture content in plat A was lower than that in plats B and C until after August 22.

Figure 10 compares the moisture content of each of the four 3-foot soil zones within each plat. It will be noted that during the early part of the period there were large differences between the moisture content in each of the four zones in plat A, smaller differences in plat B, and still smaller differences in plat C. That the moisture content of the first 3-foot zone remained the highest in all of the plats during the major part of the period is apparently due in part to differences in soil texture. Moisture equivalent determinations in each of the upper 12 feet for each plat show that the soil becomes somewhat coarser in texture with the depth.

It has commonly been assumed,[11,19] that the moisture content of the soil a few days after irrigation represented the moisture capacity and that but little moisture movement occurred after the relatively rapid movement of the first few days. Figures 8, 9, and 10 show the difficulty

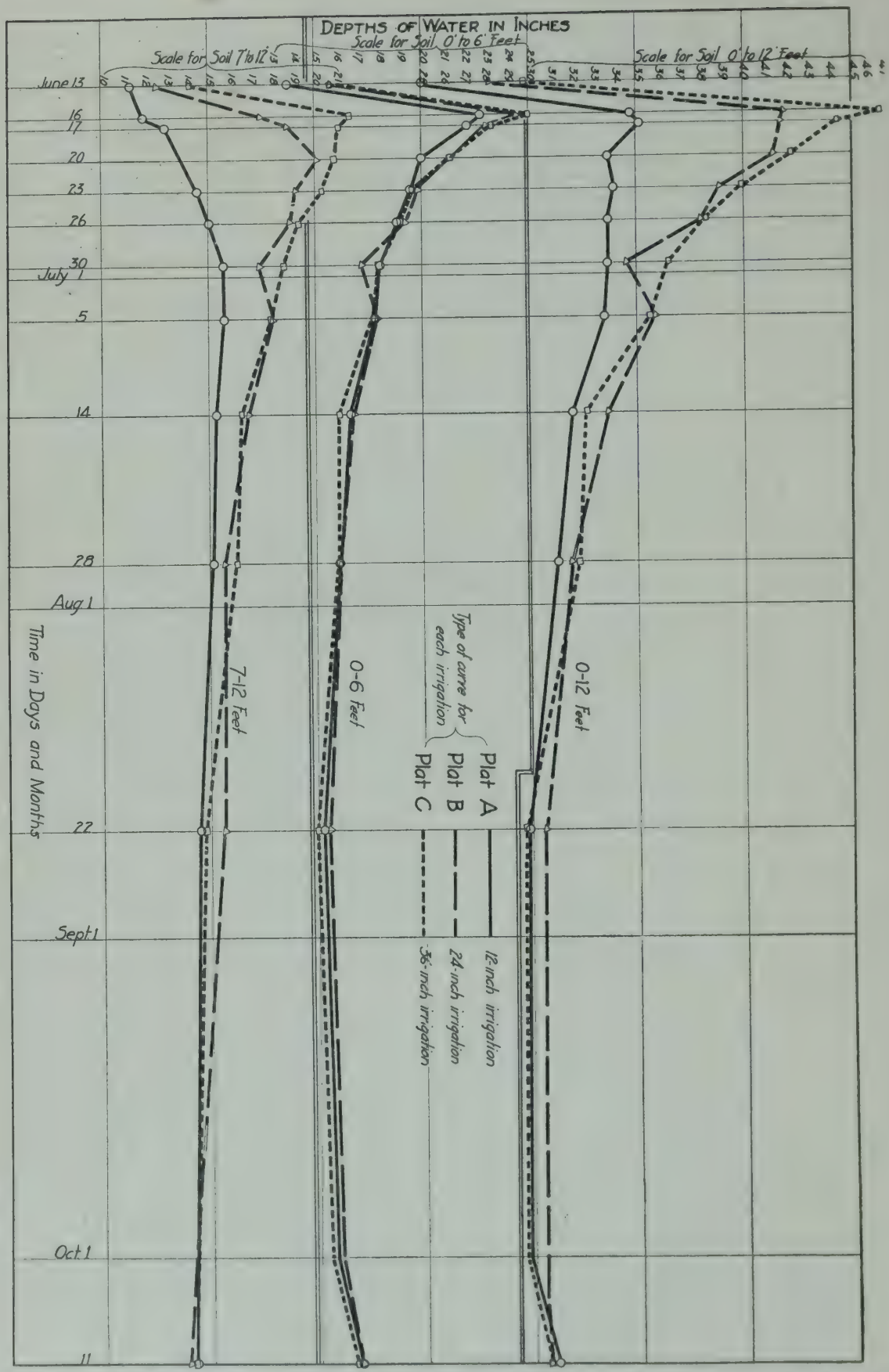

Fig. 8. Curves comparing the time rate of change in the amounts of water contained in the same depths of soil after the application of three different amounts of irrigation water. Results are expressed in inches depth of water in each of the three depths of soil considered.

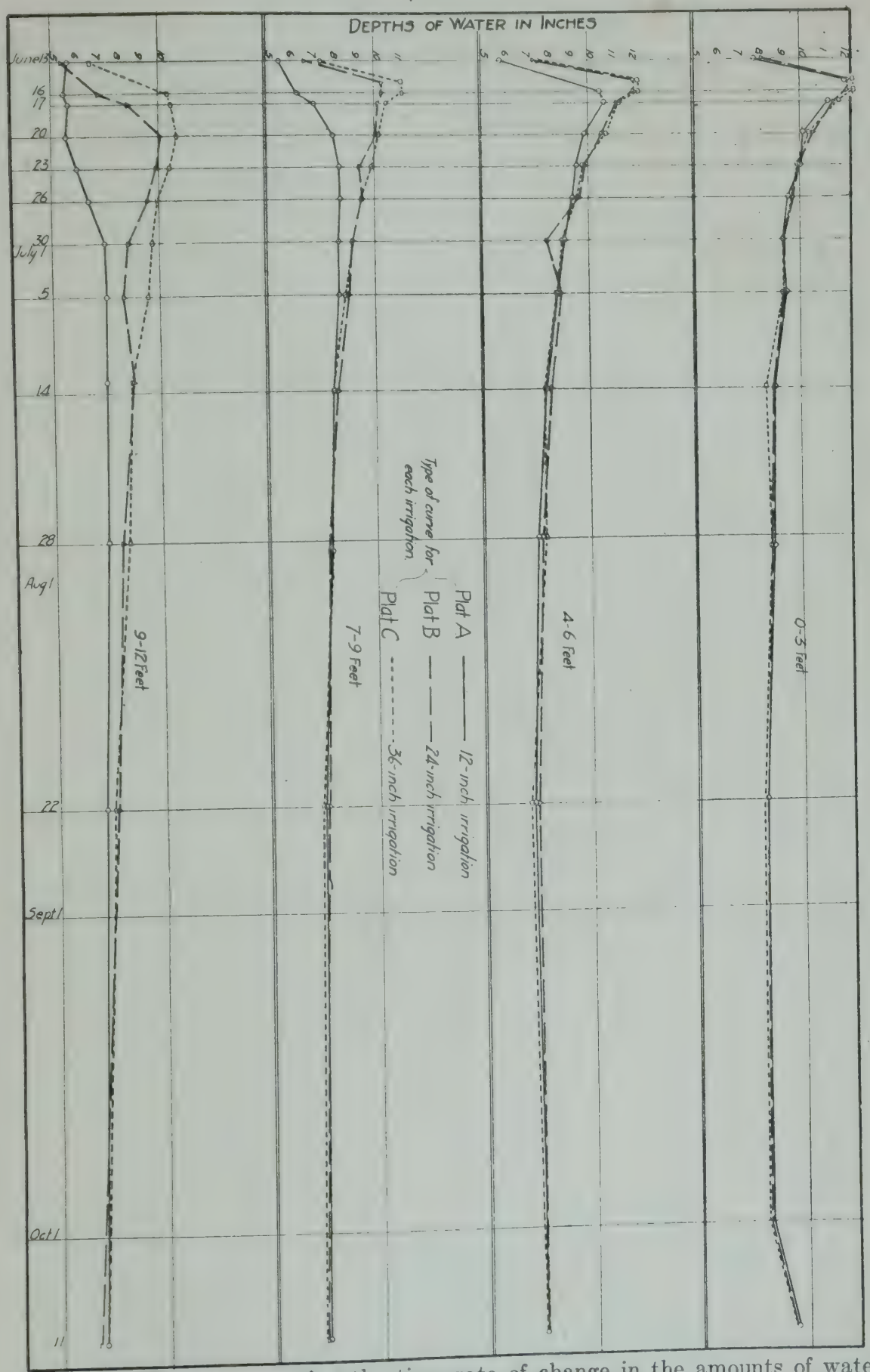

Fig. 9. Curves comparing the time rate of change in the amounts of water contained in the same depths of soil after each of three different irrigations. Results are expressed in inches depth of water in each of the four depths of soil considered.

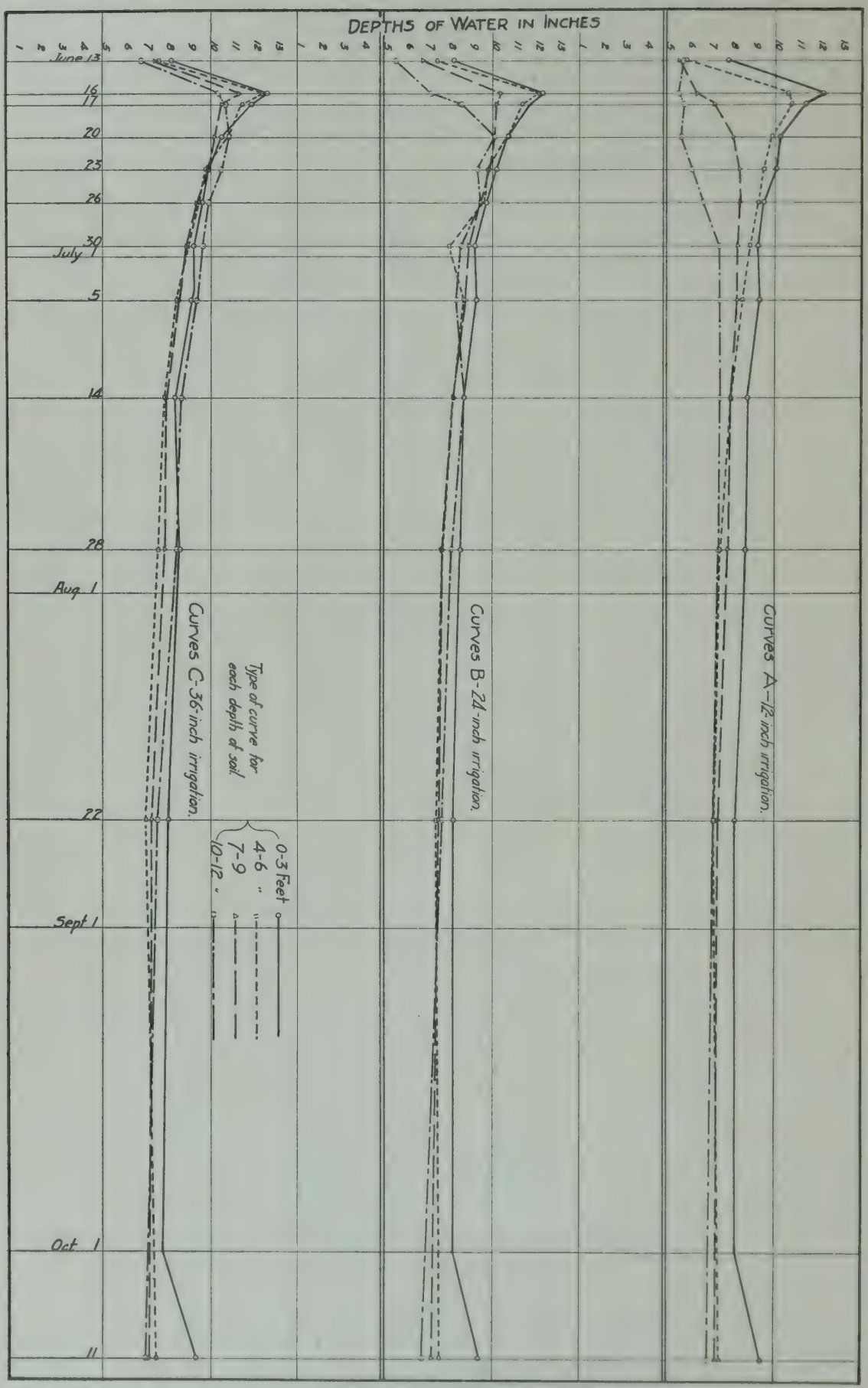

Fig. 10. Curves comparing the time rate of change in the amounts of water contained in four different depths of soil, after the application of three different amounts of irrigation water. Results are expressed in inches of water in each three feet of soil.

of selecting a particular time after irrigation as representing the maximum capacity. August 22 is apparently the only date that may be selected as representing the maximum without being arbitrary and even this date may not be chosen without the support of a mathematical analysis of all the experimental data. Such an analysis is reported after considering the vertical distribution of moisture in the soil of the three plats at different times after irrigation as shown in figures 11, 12, and 13. The moisture percentages are plotted in the fourth quadrant with the positive abscissa representing moisture content and the negative ordinate representing depth of soil in feet below the surface. In each of the figures there are five curves: (a) representing the moisture content before irrigation, (b), (c), and (d) representing the moisture content at different periods after irrigation, and (e) representing the Briggs-McLane moisture equivalent.

A comparison of the curves (a) and (b) in each of the three figures shows the influence of the amount of water applied to the soil on the depth of penetration shortly after irrigation. Curve (b) of figure 11 shows that only the upper 4.5 feet of soil had been fully moistened two days after the 12 inches of water had been applied. In figures 12 and 13 curves (c) indicate that the twelfth foot of soil was fully moistened 5 days after irrigation, at which time it contained much more moisture than it did 68 days after. In figure 11, on the other hand, the soil below a depth of 8.5 feet held appreciably more water 69 days after irrigation than it did 2 days after. Curves (d) show that 68 days after irrigation each of the plats held approximately the same amounts of moisture. Moreover, in the upper 6 feet of plats B and C the soil held no more moisture 68 days after irrigation than it did before.

Remembering that evaporation losses were reduced to a minimum and noting the heavy loss in moisture from June 16 to August 22 by downward flow it is of interest to inquire if the moisture content had reached equilibrium on August 22.

To do this Pearson's method of moments was employed to evaluate the constants in the equation

$$\rho' = \rho'_E + Ae^{-Bt} \quad \text{.....(45)}$$

in which

ρ' = the moisture percentage at a given depth of soil at any time t.

ρ'_E = the moisture percentage at equilibrium at any depth.

e = the base of natural logarithms.

t = the average time in days measured from the date of first sampling after irrigation.

A and B are constants to be evaluated from the results of the moisture determinations at different dates.

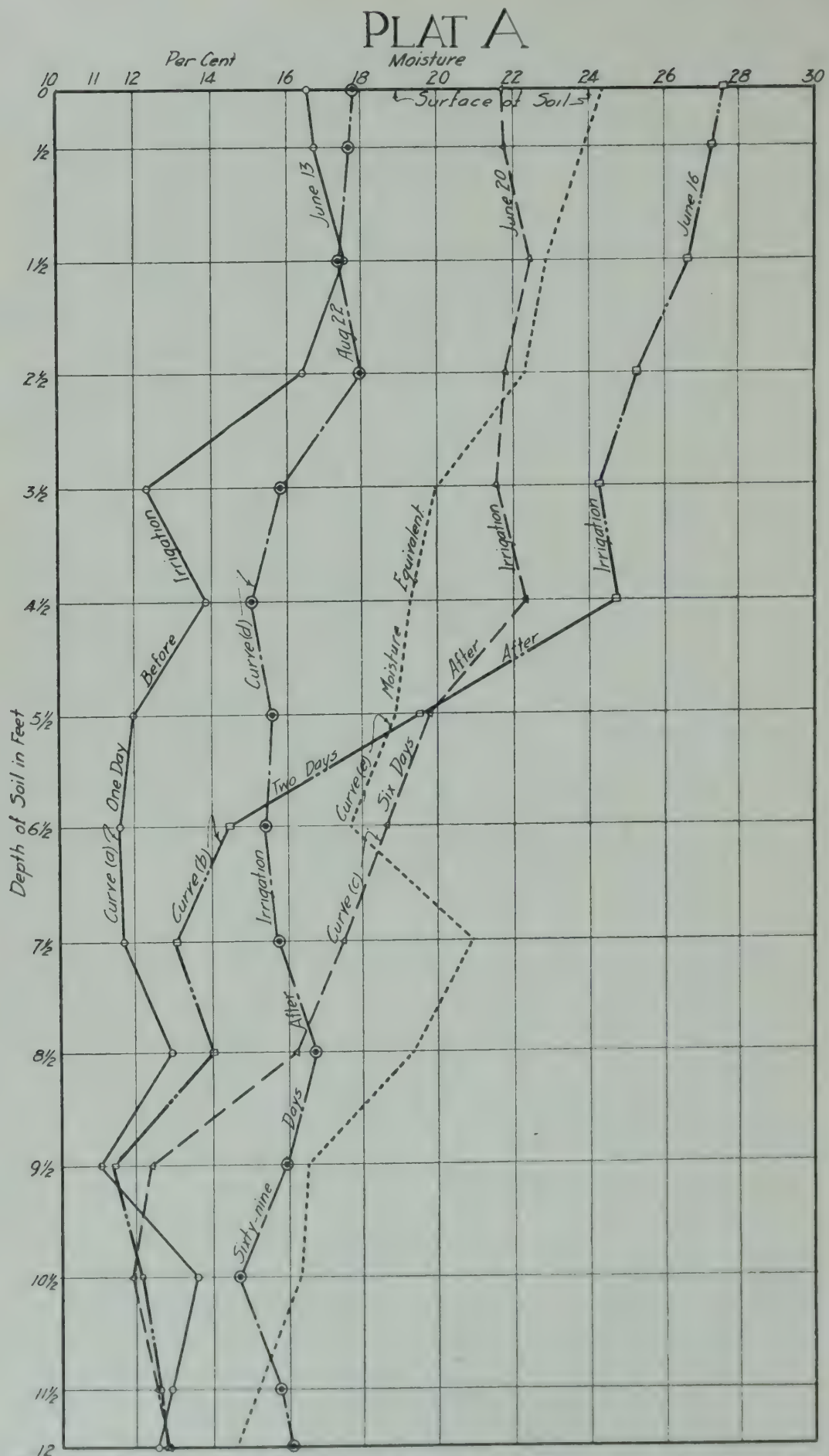

Fig. 11. Distribution of moisture in soil at different periods after a 12-inch irrigation.

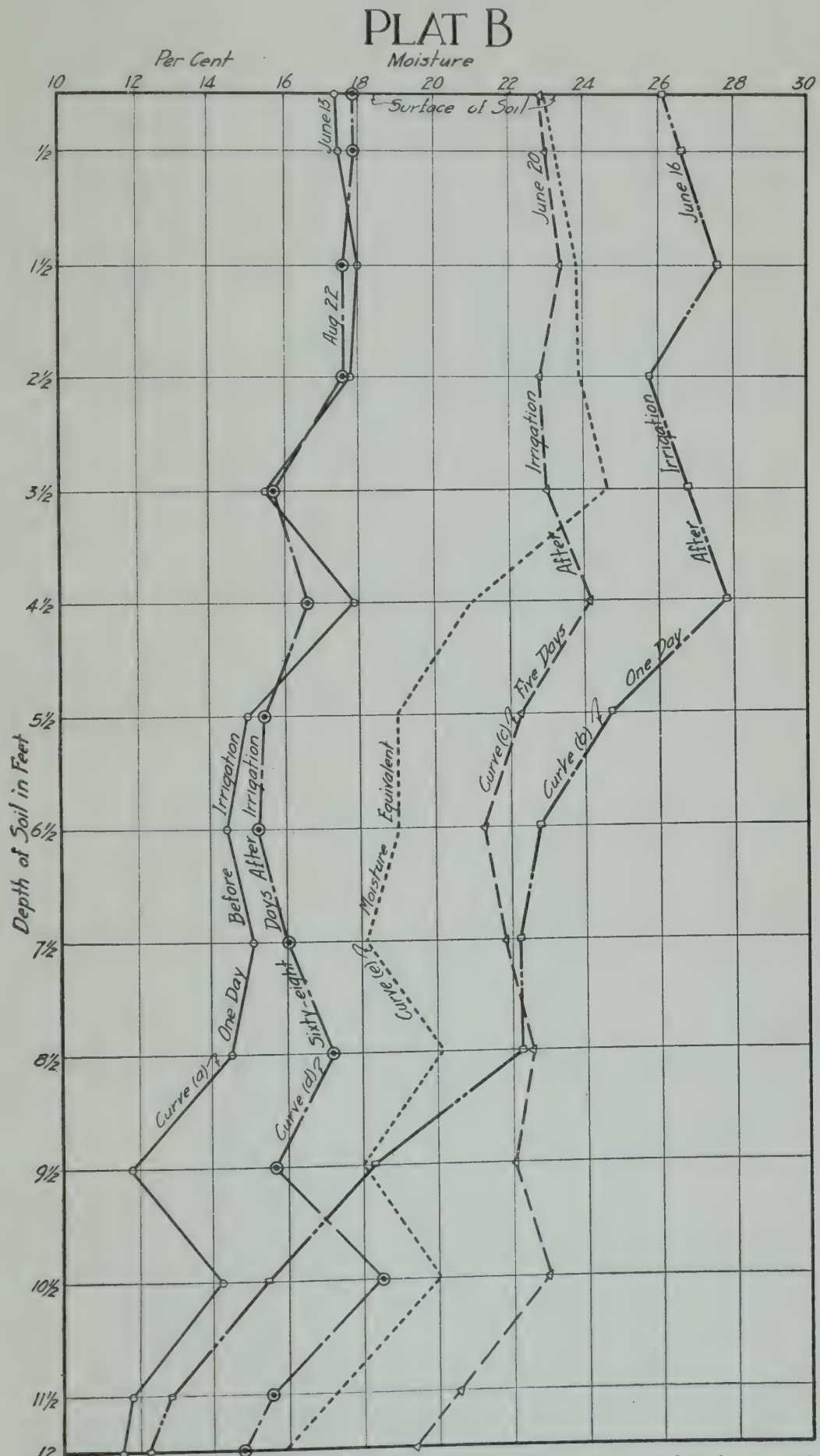

Fig. 12. Distribution of moisture in soil at different periods after a 24-inch irrigation.

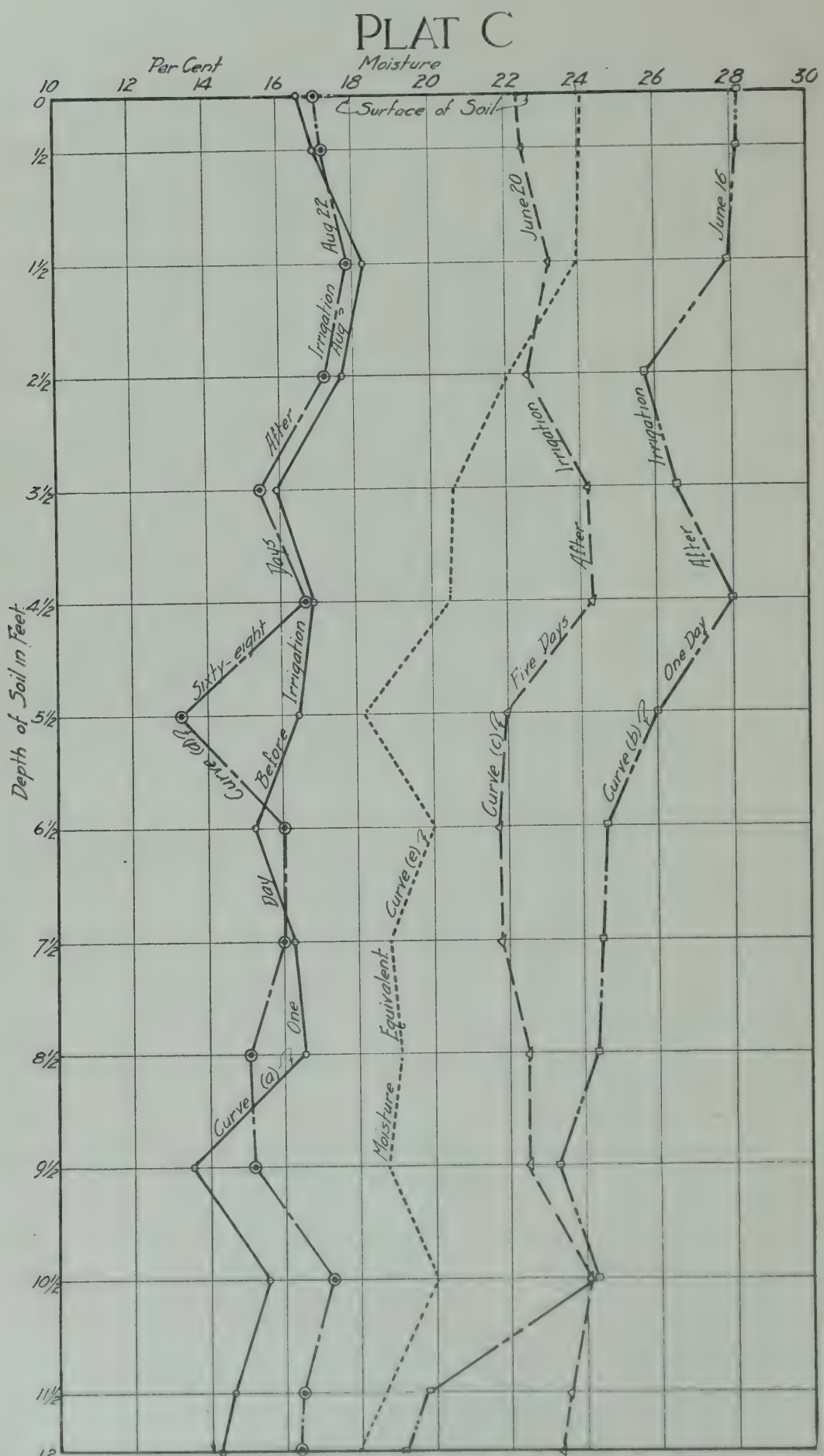
PLAT C
Per Cent Moisture
10 12 14 16 18 20 22 24 26 28 30
0 ½ 1½ 2½ 3½ 4½ 5½ 6½ 7½ 8½ 9½ 10½ 11½ 12
Depth of Soil in Feet
Surface of Soil
Curve (a) Sixty-eight Days After Irrigation Aug 22
Curve (b) One Day After Irrigation June 16
Curve (c) Five Days After Irrigation June 20
One Day Before Irrigation Aug 23
Curve (a) One Day Before Irrigation
Curve (b) One Day After Irrigation
Curve (c) Five Days After Irrigation
Moisture Equivalent Curve (c)

Fig. 13. Distribution of moisture in soil at different periods after a 36-inch irrigation.

Equations of the general type of (45) have been found to express physical relations in many natural phenomena. This type of equation seems to apply to the rate of moisture loss. According to (45), when time is infinite $\rho' = \rho'_E$, which means that the moisture in a field soil from which evaporation is prevented and in which no crops are growing in time will reach a condition of equilibrium. Finding the value of ρ'_E should, therefore, give the moisture percentages at equilibrium at the several depths of soil. It is also evident from (45) that when t equals zero $\rho' = \rho'_E + A$. The analysis by the method of moments was based on the samplings from June 16 to July 14.* A comparison of the magnitude of the moisture content at approximate equilibrium in the upper 9 feet of soil in plat C, as determined by equation (45), to the moisture content on August 22 is given below:†

Depth in feet.....	0.5	1.5	2.5	3.5	4.5	5.5	6.5	7.5	8.5
Moisture content at equilibrium ρ'_E computed.....	17.3	18.1	17.8	16.7	17.6	14.2	17.8	15.7	12.7
Moisture content on ρ' August 22.....	16.9	17.8	17.2	15.5	16.6	13.3	16.2	16.0	15.0

The differences between ρ'_E and the ρ' determined on August 22 are probably not larger than the experimental error. The analysis by which the determinations of ρ'_E were made was based on only 7 sets of moisture determinations and hence should not be interpreted as definitely establishing the equilibrium of moisture content. Since, however, the decrease of ρ' between July 28 and August 22 was comparatively small as shown in figures 8, 9, and 10, it is likely that the moisture content on the latter date closely approached the equilibrium. Had the field capillary potentiometer been perfected at the time these water-capacity experiments were conducted it would have been possible to obtain more direct and more dependable information concerning the moisture condition at equilibrium.

If the August 22 determinations do represent the moisture content at equilibrium, then ρ'_E should increase with depth of soil provided the soil were uniform in texture. The moisture equivalent determinations in each plat to a depth of 12 feet indicate a significant increase in coarse-

* The samplings of July 28 and August 22 were omitted because the large period of time between these samplings gave them undue weight in the analysis.

† The influence of soil texture on the moisture content at equilibrium with change of depth is considered in the following pages.

ness of texture with depth. The influence of this variation in texture has been eliminated to a large extent by the following procedure:

Let $\rho'_1, \rho'_2, \cdots \rho'_{12}$ be the actual moisture percentages in the respective depths on August 22.

Let $e_1, e_2, \cdots e_{12}$ be the moisture equivalent in the respective depths.

Let e_m be the mean moisture equivalent for the 12 feet.

Let $\rho'_{c1}, \rho'_{c2} \cdots \rho'_{c12}$ be the corrected moisture percentages in the respective depths on August 22.

Then

$$\rho'_{c1} = \rho'_1 \frac{e_m}{e_1}; \quad \rho_{c2} = \rho'_2 \frac{e_m}{e_2} \cdots\cdots\cdots \rho'_{c12} = \rho'_{12} \frac{e_m}{e_{12}}$$

The corrected moisture percentages for each of plats A, B, and C on August 22 are plotted in figure 14, the points for plat A being represented by circles, for plat B by triangles and those for plat C by squares. The curve of figure 14 represents the average corrected ρ'_E for the three plats. It is evident from this figure that, had the Greenville soil been uniform in texture to a depth of 12 feet, the moisture percentages 68 days after irrigation would have increased significantly with increase in depth. It is important to note that the distribution of the moisture with depth, as shown in figure 14, represents a condition that might have been qualitatively predicted from the capillary potential analysis, namely, that as the depth increases from the surface of the soil toward the water table, the numerical magnitude of the capillary potential decreases and consequently the moisture percentage of the soil must increase. The capillary potential at the surface of the plats A, B, and C cannot be computed with precision since the distance from the surface to the water table is unknown. However, letting Ψ_a be the capillary potential ½ foot below the surface where the soil samples were taken to represent the first foot and Ψ_b be the capillary potential 11½ feet below the surface, where the samples were taken to represent the 12th foot, the difference in capillary potential at the two points, according to equation (9a) is

$$\Psi_a - \Psi_b = -(1 \times 11 \times 30.5) \cdots\cdots\rightarrow = -335.5 \text{ gm.-cm. per gm.}^*$$

This capillary potential difference was accompanied by a variation in the moisture content of 4 per cent, which is the decrease in moisture between the 11.5-foot depth and the 0.5-foot depth. Assuming that

* Assuming the water table to be 60 feet below the soil surface, which is the depth to water in a neighboring well, it would follow that:

$\Psi_a = -59.5 \times 30.5 = -1815$ gm.-cm. per gm.

$\Psi_b = -48.5 \times 30.5 = -1479$ gm.-cm. per gm.

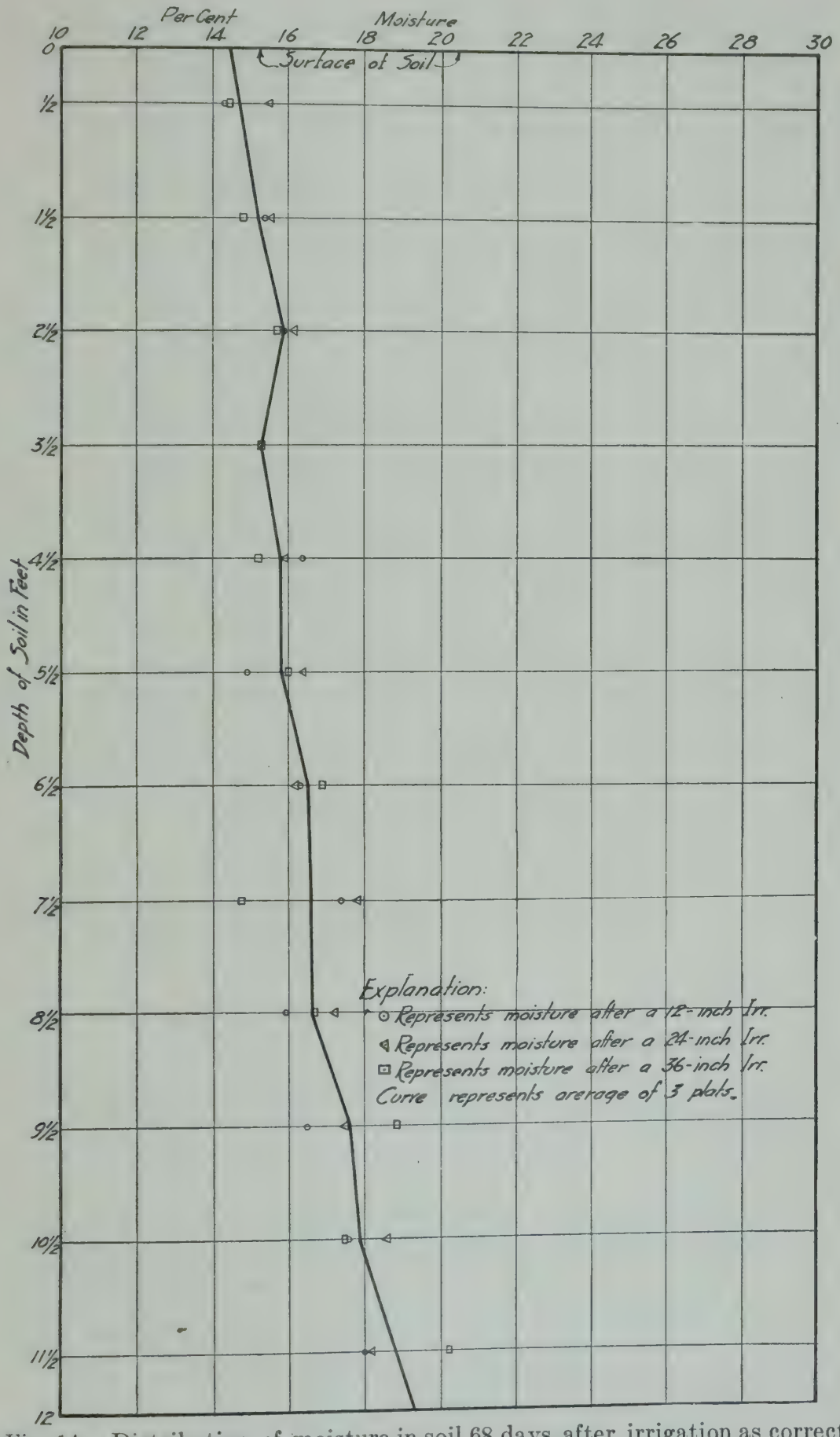

Fig. 14. Distribution of moisture in soil 68 days after irrigation as corrected for variation in soil texture, thus showing distribution that would have occurred in a soil of uniform texture.

the moisture content had reached equilibrium by this date, which was 68 days after irrigation, it is evident from the influence of change in the capillary potential on the equilibrium of the moisture content of the soil, as indicated in figures 7 and 14, that the depth to the water table influences appreciably the water capacity of the soil at equilibrium. Moreover, the recognition of the equilibrium of moisture content as an equipotential region (capillary and gravitational) is of itself sufficient basis for correcting the common belief[12,16] that the distribution of water in vertical soil columns at equilibrium is uniform. The theoretical basis for correction of this erroneous view is here supported by a study of the Greenville loam soil in the laboratory and in the field. Recent laboratory experiments by McLaughlin[16] also support the results of the above analysis.

A similar equilibrium soil moisture distribution will occur in a field of force other than the gravitational one. This has been experimentally verified[18] for a "centrifugal" force field in the laboratory of the Division of Irrigation Investigations and Practice of the University of California, Agricultural Experiment Station at Davis. Attention is directed particularly to figures 4 and 8b of the California publication reporting the above experiments, together with the accompanying discussions.

Other applications.—It is desirable briefly to view the advantages, in a study of the moisture conditions above a high water table, which result from capillary potential measurements. Referring to figure 15, measurements of the capillary potential at the points P_1, $P_2, \cdots P_n$ at elevations h_1, $h_2 \cdots h_n$ above the water table are represented by Ψ_1, $\Psi_2 \cdots \Psi_n$. If h_1, $h_2 \cdots h_n$ are measured in centimeters, they are numerically equal to the gravitational potentials φ_1, $\varphi_2 \cdots \varphi_n$ at the respective points. Suppose that $\Psi_1+h_1=\Psi_2+h_2=\cdots\Psi_n+h_n=$ constant. It would follow that the moisture distribution was in static equilibrium as represented in curve (a) of figure 15. Further, suppose that $\Psi_1+h_1>\Psi_2+h_2>\cdots\Psi_n+h_n\cdots$;* it then follows that the moisture distribution would be illustrated by curve (c) of figure 15 and that the moisture would move upward until an equilibrium of moisture distribution was reached. On the other hand, if $\Psi_1+h_1<\Psi_2+h_2<\cdots\Psi_n+h_n$, the moisture distribution would be represented by curve (b) of figure 15 and the moisture would move into the water table until equilibrium was established. Since there is no positive hydrostatic pressure above a water table it is evident that for vertical flow $\nabla\Phi$ of equation (40) would equal $\frac{(\Psi_1+h_1)-(\Psi_2+h_2)}{h_2-h_1}$ or $\frac{(\Psi_2+h_2)-(\Psi_3+h_3)}{h_3-h_2}$, etc. Hence, for any soil, the

* It is important here to remember that Ψ is negative and hence that a high numerical value results in a low absolute value.

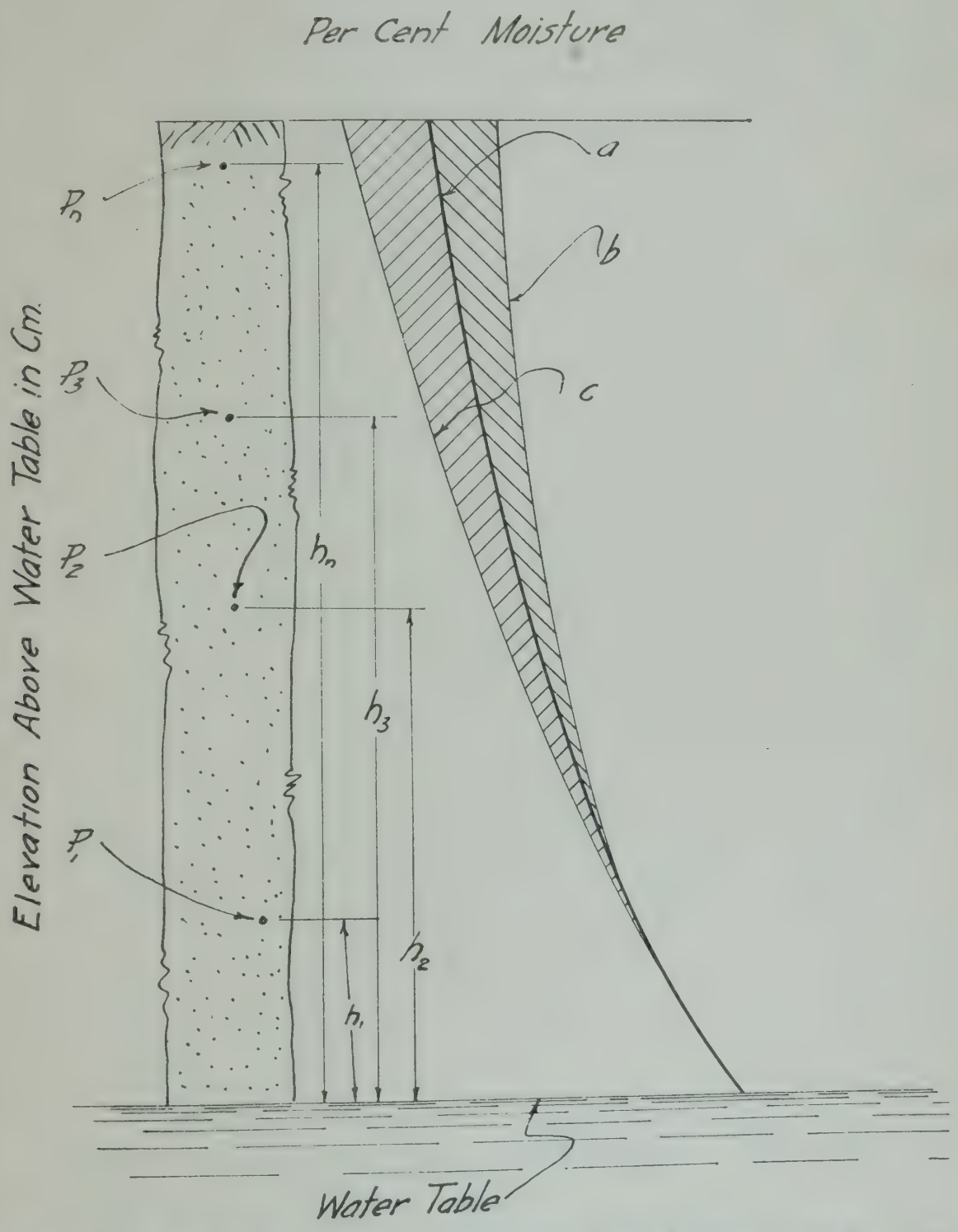

Fig. 15. The probable moisture distribution above a water table for three conditions, namely: *a*, equilibrium, in which the sum of potentials is constant, no moisture movement; *b*, Ψ is less than for equilibrium and moisture moves into water table; *c*, Ψ is greater than for equilibrium and moisture moves out of water table.

average capillary conductivity of which is known, a few measurements of capillary potential and of the average moisture content would make it possible quantitatively to determine, by use of equation (40), after a steady flow had been established, the loss from, or the contribution

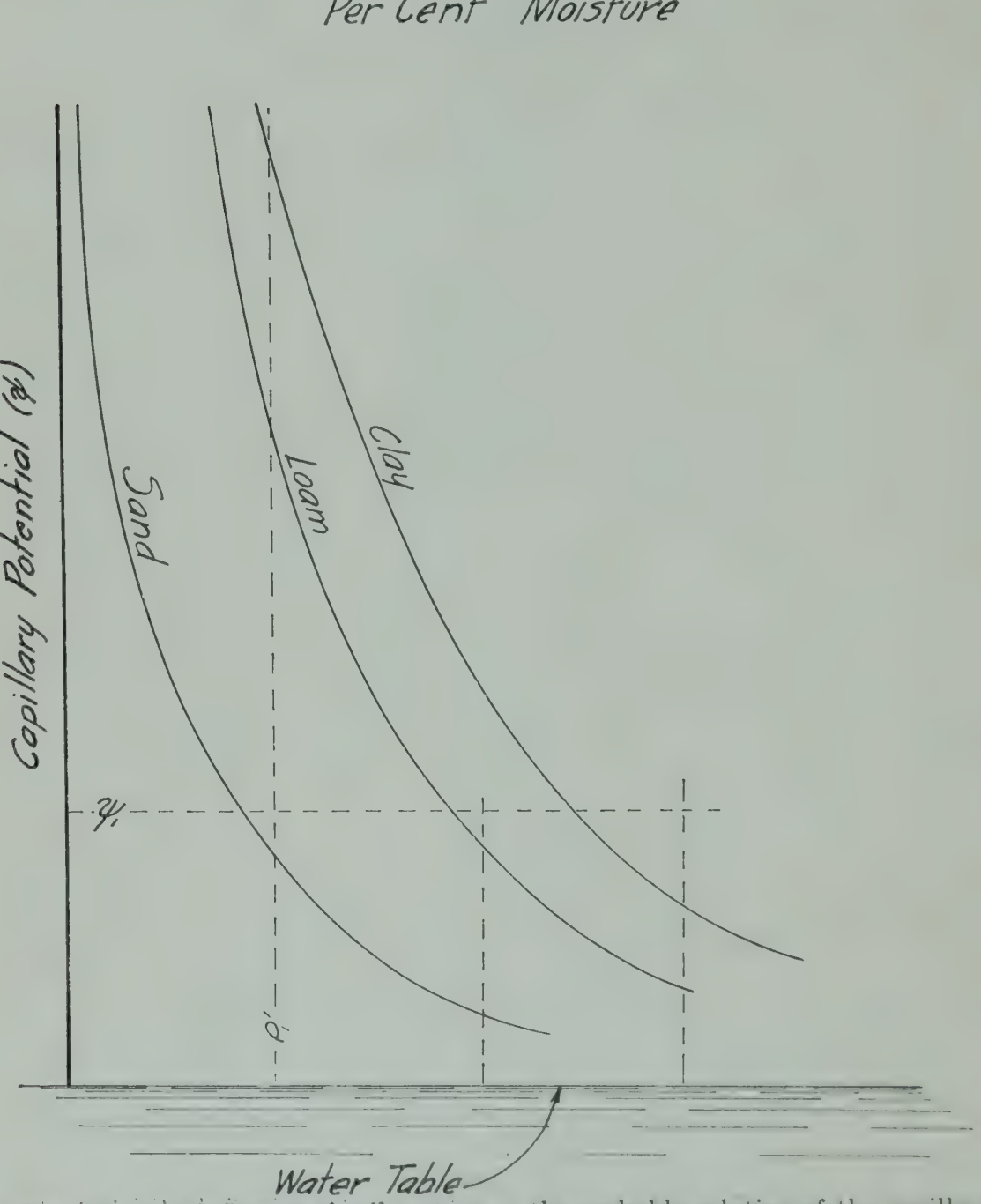

Fig. 16. The influence of soil texture on the probable relation of the capillary potential (Ψ) to the moisture content (ρ').

to, a water table resulting from the capillary stream, in a given period of time.

It appears that the quantitative relation of Ψ to ρ' is different for different types of soil. Work thus far at the Utah Agricultural Experiment Station indicates that for a given capillary potential Ψ the moisture

content is highest in a clay, medium in a loam, and lowest in a sand and, conversely, for a given ρ' the capillary potential Ψ is highest in a clay, medium in a loam, and lowest in a sand. These relations are illustrated in figure 16.

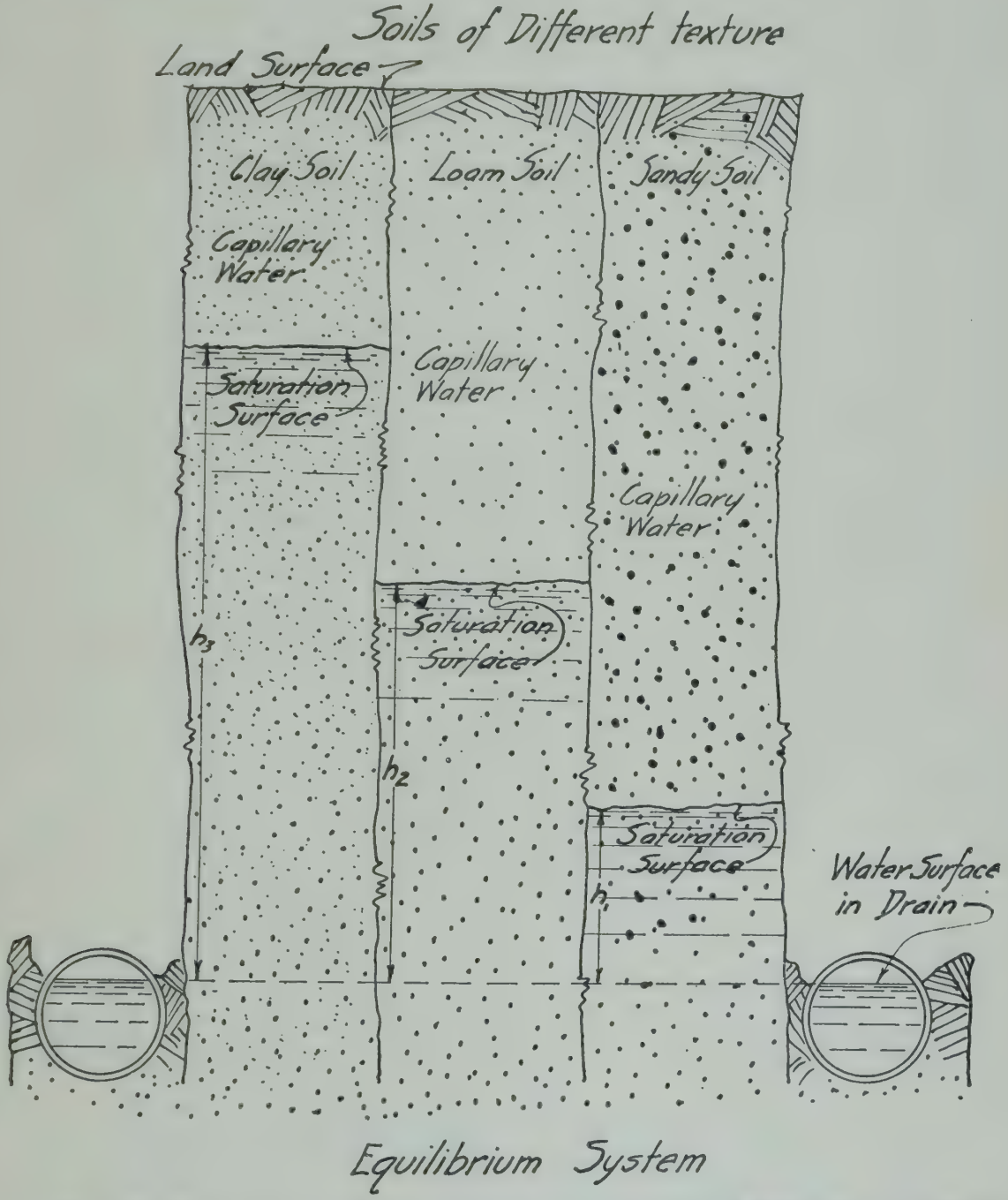

Fig. 17. The influence of soil texture on the probable differences in elevations of the water table and the surfaces of water saturation in the soil under equilibrium conditions.

The above facts have an important bearing on drainage. In the finer textured soils it is necessary to place drains comparatively deep in order to be sure that, in the upper few feet of soil, the capillary potential will be high and thus bring about a low moisture content. Moreover, both observations and experiment tend to show that very fine-textured soils

have such a high capillary potential with comparatively large percentages of capillary water that the soils actually remain saturated considerable distances above the water table as illustrated in figure 17. A very fine-textured soil thus holds the gravitational water in equilibrium above the water table a distance h_3; a medium-textured one a distance h_2; and a coarse-textured one a very small distance h_1, in which $h_3 > h_2 > h_1$.

The above examples suggest other possible applications of hydromechanics to irrigation and drainage problems. Recent applications by Linford[14] in a study of the relation of light to soil moisture phenomena seem to be very promising. The application of analytical methods to problems in irrigation and drainage, which are admittedly complex, is undoubtedly a more rational procedure than the pursuing of empirical methods without the guidance of fundamental principles. The heterogeneity of soils in reality increases rather than decreases the need for applying the laws of motion to these problems, despite the conditions of extreme variability sometimes encountered in soils in which measurements of soil moisture content or flow by any method are impracticable.

SUMMARY AND CONCLUSIONS

1. Knowledge of the laws which govern the movement of ground water and soil moisture is essential to its effective control.

2. The fundamental hydrodynamical equation of motion and the equation of continuity may be applied to irrigation and drainage problems.

3. For irrotational motion the velocity may be derived from a potential.

4. The driving forces in the formula for flow of water in open channels and in pipes are derivable from a potential.

5. Applications of hydrodynamics to ground-water and soil-moisture movement have been made by only a few investigators.

6. The capillary potential can be measured in the laboratory with the aid of porous cups by methods herein described.

7. The capillary potential Ψ is a function of the moisture content ρ'. Analysis of 84 determinations of the relation of Ψ to ρ' suggest that it may be represented by an equilateral hyperbola of the form $(\rho' - a)(\Psi + b) = c$.

8. The moisture distribution at equilibrium in a vertical soil column is not uniform but decreases with height above the water table. Such distribution qualitatively confirms the requirements for an equipotential moisture region.

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LITERATURE CITED

[1] Briggs, Lyman J.
1899. The movement and retention of water in soils. U. S. Dept. Agr. Yearbook 1898: 399–404.

[2] Briggs, Lyman J.
1898. The mechanics of soil moisture. U. S. Dept. Agr. Bur. Soils Bul. **10**:1–24. figs. 1–7.

[3] Buckingham, Edgar
1907. Studies on the movement of soil moisture. U. S. Dept. Agr. Bur. Soils Bul. **38**:1–61. figs. 1–23.

[4] Coffin, Joseph George
1911. Vector analysis. 1–253. figs. 1–69. John Wiley & Sons, N. Y.

[5] Etcheverry, B. A.
1915. Irrigation practice and engineering—Conveyance of water. **2**:1–350. figs. 1–81. McGraw-Hill Book Co., N. Y.

[6] Gardner, Willard
1919. The movement of moisture in soil by capillarity. Soil Sci. **7**:314–315. figs. 1–3.

[7] Gardner, Willard
1920. The capillary potential and its relation to soil-moisture constants. Soil Sci. **10**:357–359. fig. 1.

[8] Gardner, Willard and John A. Widtsoe.
1921. The movement of soil moisture. Soil Sci. **11**:215–232. figs. 1–11.

[9] Houstoun, R. A.
1920. An introduction to mathematical physics. 1–197. figs. 1–75. Longmans, Green & Co., N. Y.

[10] Israelsen, O. W.
1918. Studies on capacities of soils for irrigation water. Jour. Agr. Res. **13**:1–36. figs. 1–14. pl. 1.

[11] Israelsen, O. W. and Frank L. West.
1922. Water-holding capacity of irrigated soils. Utah Agr. Exp. Sta. Bul. **183**:1–24. figs. 1–7.

[12] Keen, B. A.
1924. On the mutual relationships in ideal soil. Jour. Agr. Sci. **14**:170–177. figs. 1–4.

[13] Lamb, Horace
1916. Hydrodynamics. Fourth edition. 1–701. Cambridge University Press, Cambridge, England.

[14] Linford, Leon B.
1926. The relation of light to soil moisture phenomena. Soil Sci. **10**:233–251. figs. 1–6.

[15] McLaughlin, W. W.
1920. Capillary movement of soil moisture. U. S. Dept. Agr. Bul. **835**:1–69. figs. 1–10.

[16] McLaughlin, W. W.
1924. The capillary distribution of moisture in soil columns of small cross section. U. S. Dept. Agr. Bul. **1221**:1–122. figs. 1–7.

[17] Slichter, Charles S.
1898. Theoretical investigations of the motion of ground water. U. S. Geol. Survey 19th Ann. Rept. **2**:301–380.

[18] Veihmeyer, F. J., O. W. Israelsen, and J. P. Conrad.
1924. The moisture equivalent as influenced by the amount of soil used in its determination. California Agr. Exp. Sta. Tech. Paper **16**:1–60. figs. 1–9. pl. 1.

[19] Widtsoe, John A. and W. W. McLaughlin.
1912. The movement of water in irrigated soils. Utah Agr. Exp. Sta. Bul. **115**:199–268. fig. 1. pls. 6.

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VARIATION IN THE REACTIONS OBTAINED IN REPEATED AGGLUTINATION TESTS OF THE SAME FOWLS WITH *BACTERIUM PULLORUM* ANTIGEN

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INTRODUCTION

The studies by Rettger[1, 2, 4] and Rettger and Harvey,[3] reported in four papers published between 1900 and 1909, definitely established the disease of young chicks commonly known as "white diarrhea" to be a specific infectious disease, the causative organism of which was designated *Bacterium pullorum*. Further studies by Rettger and his associates were reported in 1909,[5] 1911[6] 1912,[7] and 1914.[8, 9] They determined that apparently healthy adult fowls may be carriers of *Bact. pullorum*. The infection in hens usually becomes localized in the ovaries and is eliminated in the eggs. When such eggs are used for hatching, the infection is transmitted to chicks. This is considered the usual source of *Bact. pullorum* infection in chicks. Jones[10, 11] in 1910 and 1911 and Gage[12] in 1911 published the results of investigations which confirmed the findings of Rettger and his associates.

The most important problem in the prevention of the disease in chicks, therefore, became the detection and elimination of infected breeding stock. In 1913 Jones[13] demonstrated that the agglutination test was of value for this purpose. His findings were confirmed by others and the testing of breeding flocks by this method has been practiced extensively for several years.

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All who have made careful study of the agglutination test for detecting carriers of *Bact. pullorum* recognize that repeated tests are necessary for the elimination of all infected fowls in a flock. The failure of a single test to detect all of the infected fowls is commonly considered to be due either to certain birds having acquired infection too recently for the production of sufficient agglutinins in their blood serum to cause an agglutination reaction or to certain birds becoming infected after the test, either from association with the infected birds or from contaminated litter and soil. It is commonly considered, however, that after agglutinins have become sufficiently abundant to cause a reaction, they will remain so as long as the fowl continues to harbor *Bact. pullorum.*

The workers in the laboratories of the Division of Veterinary Science, University of California had no reason to doubt that fowls with well-established infection with *Bact. pullorum* would uniformly react to the agglutination test until, in the routine testing that was being carried on, it became necessary to make tests of two lots of blood samples from the same birds at an interval of twenty-one days. On August 25, 1925, a test was made on a lot of 574 blood samples. Another lot of blood samples from this flock was received on September 15, 1925, and included duplicates of 388 samples that had been tested twenty-one days previously. The comparative results of the tests of the two samples of blood from 388 fowls were as follows:

Six fowls reacted to both tests.

Twelve fowls that gave a positive reaction to the first test failed to react to the second test.

Three fowls that were negative at the first test reacted to the second test.

Such discrepancies in the results of the two tests could not be ascribed to differences between the antigens or methods because these were the same for both tests. They could hardly be considered as due to certain fowls having become free from and others having acquired infection between the two tests because of the short interval between them. Therefore, it seemed probable that all of the fowls that had reacted to either one or both tests were infected at the time each test was made but certain of them had failed to react to one test. If this assumption is correct, it would indicate that fowls may not constantly give a positive reaction to the agglutination test while they are carriers of *Bact. pullorum.* It was to obtain information on this point that the studies herein reported were undertaken.

Other points regarding which it was thought information might be obtained were:

First, the correlation between intensity of egg production, age of the birds or season of the year and any variability in the reactions to repeated agglutination tests of the same individuals that is found to occur;

Second, the accuracy with which the results of an agglutination test may be interpreted to indicate the presence or absence of *Bact. pullorum* infection;

Third, the rapidity with which *Bact. pullorum* infection may spread among non-infected adult females from association with infected adult females.

Plan of the Experiment

In December, 1925, 200 White Leghorn pullets, from 8 to 9 months old, were obtained from a flock in which *Bact. pullorum* infection was known to exist. They were immediately leg-banded and subjected to an agglutination test. Seventy of the birds gave a positive reaction in at least one dilution and 130 failed to react. The seventy reactors and eighty of the non-reactors were placed together in one house and designated group 1 and group 2, respectively. The remaining fifty non-reactors were placed in a separate house and designated group 3. Outside runs were not provided. An agglutination test of the blood serum of all birds was made once each month. This procedure is to continue for at least two years, but this report is concerned only with the first twelve months.

A careful search for *Bact. pullorum* was made in all birds that died from any cause, except certain ones otherwise accounted for.

The antigen was prepared from a single strain of *Bact. pullorum* of known good agglutinability that had been isolated from a chick. Cultures were incubated on agar for 48 hours and the growth washed off with sterile, physiological salt solution containing 0.5 per cent phenol. Cultural and microscopic tests were made of each lot of antigen to insure freedom from contamination. The antigen in concentrated form was stored in an ice box. It was diluted with sufficient phenolized saline to give a reading of 3.5 cm. with a Gates' nephelometer at the time the tests were made.

In all of the tests, four dilutions of serum and antigen were used, i.e., 1–25, 1–50, 1–100 and 1–200. The tubes were incubated for 24 hours and kept at room temperature 24 hours longer. Readings were made after 24 and 48 hours.

Summary and Discussion of the Results of the Agglutination Tests of the Fowls in Group 1

This group consisted of seventy birds that gave a positive reaction to the first agglutination test. However, there was a marked variation in the number of these birds that reacted at each of the eleven subsequent monthly tests. A summary of the results of the tests and the average egg production during each month is given in table 1. The gradual decrease in the number of birds in the group is due to deaths that occurred.

TABLE 1

Summary of the Results of 12 Agglutination Tests and Egg Production of Group 1

Month	Number of birds in group	Number of reactors	Per cent reactors	Per cent egg production for the month
December	70	70	100.0	0.
January	70	38	54.2	1.3
February	68	39	57.3	2.9
March	60	44	73.3	27.2
April	59	29	49.1	39.5
May	58	26	44.8	45.7
June	56	23	41.0	41.4
July	54	31	57.3	28.2
August	53	26	49.0	21.7
September	51	25	49.0	21.9
October	51	22	43.1	12.2
November	50	19	38.0	3.6

In table 1, it is seen that at none of the tests after the first were reactions obtained from all of the birds that reacted to the first test. The nearest approach to this was in March, when 73.3 per cent of the birds reacted. In January, February and July, positive reactions were obtained from 54.2 per cent, 57.3 per cent and 57.3 per cent of the birds, respectively. Less than half of the birds gave a positive reaction at each of the seven other tests, the percentage ranging from 49 in April, August and September down to 38 in November. Table 1 also clearly shows that the variation in the number of birds that reacted at the different tests was not correlated to that of egg production.

The progressive decrease in the number of reactors to each of the tests after July suggests that the decrease may be in correlation with

the increasing age of the birds. However, a similar decline in number of reactors occurred between March and June, but was followed by an increase in the number of reactors to the test in July.

The difference in the number of the birds of group 1 that reacted to each of the twelve tests was not merely a progressive decrease due to certain of the birds ceasing to react, but was also due to a fluctuation between positive and negative of the reactions which some individual birds gave to the different agglutination tests. This is shown by the increase in the number of positive reactions obtained in March over that obtained in January or February and in the number in July over that obtained in April, May or June. It is more clearly brought out, however, by the following detailed summary of the reactions to the agglutination test of the fifty birds that lived during the entire year and were tested twelve times.

A general summary of the number of the fifty birds that reacted to each of the twelve tests is given in table 2.

TABLE 2

SUMMARY OF RESULTS OF AGGLUTINATION TESTS OF 50 BIRDS OF GROUP 1 THAT WERE TESTED TWELVE TIMES

Number of test	Month	Number of reactors	Per cent reactors
1	December	50	100.0
2	January	31	62.0
3	February	28	56.0
4	March	36	72.0
5	April	23	46.0
6	May	21	42.0
7	June	19	38.0
8	July	27	54.0
9	August	24	48.0
10	September	24	48.0
11	October	21	42.0
12	November	19	38.0

In table 2, it is seen that the variation in the percentage of the fifty birds that gave positive reactions to the different agglutination tests closely follows that shown in table 1 for the whole of group 1.

Of the fifty birds that were tested twelve times

10, or 20 per cent gave a positive reaction to all 12 tests.
4, or 8 per cent gave a positive reaction to 11 tests.
4, or 8 per cent gave a positive reaction to 10 tests.
2, or 4 per cent gave a positive reaction to 9 tests.

1, or 2 per cent gave a positive reaction to 8 tests.
3, or 6 per cent gave a positive reaction to 7 tests.
1, or 2 per cent gave a positive reaction to 6 tests.
4, or 8 per cent gave a positive reaction to 5 tests.
4, or 8 per cent gave a positive reaction to 4 tests.
5, or 10 per cent gave a positive rtaction to 3 tests.
4, or 8 per cent gave a positive reaction to 2 tests.
8, or 16 per cent did not react after the first test.

The distribution of the positive and negative reactions to the agglutination tests of the forty birds that did not give a positive reaction to all of the twelve tests is given in table 3.

A study of table 3 shows that the positive reactions of twenty-six of the thirty-two fowls that reacted to from two to eleven tests were interspersed with negative reactions to one or more consecutive tests. The most commonly occurring irregularity of this nature was one negative reaction between two positive reactions. This occurred in nineteen instances. Negative reactions to two consecutive tests between positive tests occurred in seven instances; to three consecutive tests in three instances; to four consecutive tests in three instances; to five consecutive tests in three instances; and to seven consecutive tests in one instance.

Table 3 also shows that certain of the birds, after giving positive reactions either consistently or irregularly to one or more tests, did not react to any subsequent test. The number of such birds and the last test to which a positive reaction was obtained is as follows:

8 birds did not react after the first test.
2 birds did not react after the third test.
3 birds did not react after the fourth test.
3 birds did not react after the fifth test.
3 birds did not react after the eighth test.
1 bird did not react after the ninth test.
5 birds did not react after the tenth test.
4 birds did not react after the eleventh test.

The disappearance of agglutinins from the blood serum of the sixteen birds that failed to react after the first, third, fourth or fifth test is possibly due to the birds having become free from *Bact. pullorum* infection. These birds cannot with certainty be regarded as free from infection, however, because, as will be shown later, *Bact. pullorum* was isolated from the ovaries of six birds of group 1 that died after having failed to react to from one to three agglutination tests next preceding their deaths.

TABLE 3

THE DISTRIBUTION OF THE POSITIVE AND NEGATIVE REACTIONS OF 40 BIRDS OF GROUP 1 THAT DID NOT REACT TO ALL OF THE TWELVE AGGLUTINATION TESTS

Number of positive reactions	Total number of birds	Number of birds that gave the same reaction to each test	Tests at which a positive reaction occurred	Tests at which a negative reaction occurred
11	4	1	First and second; fourth to twelfth	Third
		1	First to sixth; eighth to twelfth	Seventh
		2	First to eleventh	Twelfth
10	4	1	First to fifth; seventh to ninth; eleventh and twelfth	Sixth and tenth
		1	First to tenth	Eleventh and twelfth
		1	First to fourth; sixth; eighth to twelfth	Fifth and seventh
		1	First to third; fifth to eleventh	Fourth and twelfth
9	2	1	First to fourth; sixth to tenth	Fifth, eleventh and twelfth
		1	First and second; sixth to twelfth	Third, fourth and fifth
8	1	1	First; third to fifth; eighth and ninth; eleventh and twelfth	Second, sixth, seventh, tenth
7	3	1	First to fourth; ninth and tenth; twelfth	Fifth to eighth; eleventh
		1	First; seventh to twelfth	Second to sixth
		1	First to sixth; eleventh	Seventh to tenth; twelfth
6	1	1	First and second; fourth to sixth; eighth	Third; seventh; ninth to twelfth
5	4	1	First to fifth	Sixth to twelfth
		1	First and second; fourth and fifth; eighth	Third; sixth and seventh; ninth to twelfth
		1	First to fourth; twelfth	Fifth to eleventh
		1	First to third; eighth and ninth	Fourth to seventh; tenth to twelfth

TABLE 3—(*Continued*)

Number of positive reactions	Total number of birds	Number of birds that gave the same reaction to each test	Tests at which a positive reaction occurred	Tests at which a negative reaction occurred
4	4	1	First and second; fourth and fifth	Third; sixth to twelfth
		1	First; fourth; eighth; tenth	Second and third; fifth to seventh; ninth; eleventh and twelfth
		1	First to fourth	Fifth to twelfth
		1	First, third and fourth; tenth	Second; fifth to ninth; eleventh and twelfth
3	5	1	First and second; fourth	Third; fifth to twelfth
		1	First; fourth; tenth	Second and third; fifth to ninth; eleventh and twelfth
		1	First; fourth; eighth	Second and third; fifth to seventh; ninth to twelfth
		1	First; fourth and fifth	Second and third; sixth to twelfth
		1	First, second and third	Fourth to twelfth
2	4	3	First and fourth	Second and third; fifth to twelfth
		1	First and third	Second; fourth to twelfth
1	8	8	First	Second to twelfth

Any or all of the birds that gave positive reactions up to the eighth or subsequent tests can reasonably be expected to again react since the number of tests to which these birds have given a negative reaction is no greater than the number of consecutive negative reactions that occurred between the positive reactions of some of the birds that reacted irregularly to the tests.

The variation in the number of birds of group 1 that reacted to each of the twelve agglutination tests made at intervals of approximately one month is, therefore, manifested in two ways: first, by fluctuation between positive and negative of the reactions of some individuals to the different tests, and, second, by certain of the birds, after giving a positive reaction to one or more tests, ceasing to react to all subsequent tests.

Studies by Beach, Halpin and Lampman[14] that were carried on at the same time as those herein reported showed similar variation in the reactions to the agglutination test exhibited by a flock of hens that was tested twelve times in thirteen months.

Summary of Results of the Agglutination Tests and Postmortem Examination of the Fowls that Died in Group 1

The mortality in group 1 during the year was twenty fowls. Two were not examined. The remaining eighteen were carefully examined for the presence of gross ovarian or other lesions suggestive of *Bact. pullorum* infection. A bacteriologic examination, particularly for the purpose of determining the presence of *Bact. pullorum,* was made of the ovaries and yolks of these birds. The results are given in table 4:

TABLE 4

Results of Agglutination Tests and Postmortem Examination of Twenty Fowls that Died in Group 1

Number of agglutination tests	Tests giving positive reaction	Tests giving negative reaction	Condition of ovary	Ovarian lesions found	Results of bacteriologic examination of ovaries
2	First	Second	Active	None	Negative
3	All	None	Dormant	Abnormal yolks	*Bact. pullorum* isolated
3	First	Second and third	Dormant	Congestion. No abnormal yolks	*Bact. pullorum* isolated
4	First and second	Third and fourth	Dormant	Abnormal yolks	*Bact. pullorum* isolated
7	First, third and fourth	Second, fifth, sixth, seventh	Not examined		
8	All	None	Active	Abnormal yolks	*Bact. pullorum* isolated
10	All	None	Dormant	Abnormal yolks	*Bact. pullorum* isolated
11	All	None	Dormant	Abnormal yolks	*Bact. pullorum* isolated
3	First	Second and third	Dormant	Abnormal yolks	*Bact. pullorum* isolated
2	First	Second	Dormant	None	*Bact. pullorum* isolated
3	First and third	Second	Dormant	Abnormal yolks	*Bact. pullorum* isolated
6	First to fifth	Sixth	Dormant	Abnormal yolks	*Bact. pullorum* isolated
3	First and third	Second	Dormant	Abnormal yolks	*Bact. pullorum* isolated
5	First, third, fourth	Second, fifth	Active	Abnormal yolks	Negative
9	First, third, fourth, sixth, eighth	Second, fifth, seventh, ninth	Active	Abnormal yolks	*Bact. pullorum* isolated
3	First	Second, third	Dormant	Abnormal yolks	*Bact. pullorum* isolated
6	First	Second to sixth	Not examined.		
8	All	None	Dormant	Abnormal yolks	*Bact. pullorum* isolated
3	First	Second, third	Dormant	Abnormal yolks	*Bact. pullorum* isolated
4	First	Second, third, fourth	Active	Abnormal yolks	*Bact. pullorum* isolated

A study of the summarized data concerning the twenty dead fowls from group 1, as given in table 4, shows the following:

No examination was made of two fowls.

Bact. pullorum was isolated from the ovaries of sixteen of the eighteen fowls examined. Gross ovarian lesions, in the form of abnormal yolks, were present in fourteen of these. Congestion of the ovary was the abnormality found in one. In the one remaining fowl from which *Bact. pullorum* was isolated, no ovarian abnormality nor other lesion suggestive of infection with *Bact. pullorum* was observed. Five of the sixteen fowls that yielded *Bact. pullorum* at the bacteriologic examination reacted to all agglutination tests before their death. Three failed to react to all tests but did to the one next preceding their death. Two did not react to the last agglutination test, five did not react to the last two tests, and one did not react to the last three tests, preceding their death. This definitely shows fowls with well-established ovarian infection with *Bact. pullorum* may not always have sufficient agglutinins in their blood serum to cause a reaction to the agglutination tests.

The ovary of one of the two reacting fowls from which *Bact. pullorum* was not isolated was normal in appearance. The ovary of the other bird contained abnormal yolks. The negative results of the bacteriologic examination of these two birds cannot be considered as positive evidence that they were not carriers of *Bact. pullorum.* The organism may have been present in them even though it was not recovered in cultures.

The comparative results of the agglutination tests and bacteriologic examination of the eighteen dead fowls indicate that a high percentage of fowls that give a positive reaction to an agglutination test for the detection of *Bact. pullorum* infection are carriers of that organism. This is true even though such fowls fail to react to agglutination tests made one to three months later. As previously stated, the term "positive reaction" in this paper is applied to partial or complete clearing of any one or all of the four serum-antigen dilutions, 1–25, 1–50, 1–100 and 1–200.

Summary of Results of the Agglutination Tests of the Fowls in Group 2

Group 2 consisted of eighty of the fowls that did not react to the first agglutination test and that were confined in the same pen with group 1, the fowls that reacted to the first test.

Twelve of these fowls reacted to some of the subsequent agglutination tests. The tests to which these birds gave positive reactions were as follows:

One fowl gave a positive reaction to the second, third, fourth and fifth tests, and a negative reaction to the sixth test. This bird afterwards disappeared from the pen and no further data concerning it was obtained.

One fowl gave a positive reaction to the fourth, fifth and eighth to twelfth tests.

One fowl gave a positive reaction to the fourth, fifth, eighth and ninth tests.

One fowl gave a positive reaction to the fourth and eighth tests.

Five fowls gave a positive reaction to the fourth test only. The reactions of two of these consisted only of a partial clearing of the lowest of the four serum-antigen dilutions. Since these two birds gave only a slight reaction to the one test and no reaction to the other tests, it is perhaps incorrect to classify them as reactors.

One fowl gave a positive reaction to the eighth and ninth tests.

One fowl gave a positive reaction to the tenth test.

One fowl gave a positive reaction to the eleventh and twelfth tests.

With the exception of the one that disappeared from the pen, all of the fowls that became reactors to the agglutination test are still living. No opportunity for postmortem and bacteriologic examination of any of them has, therefore, been afforded.

Nine of the fowls were negative to from one to three agglutination tests before they gave a positive reaction. This number of negative reactions is no greater than the number of consecutive negative reactions that occurred between the positive reactions of some of the birds of group 1. Therefore, it seems just as probable that the positive reactions of these birds resulted from *Bact. pullorum* infection which they were harboring when the experiment started as from infection which they acquired from association with the infected birds.

The other three birds that became reactors did not react until the eighth, tenth and eleventh tests, respectively. It does not seem improbable, therefore, that the positive reaction to the agglutination test of these birds was due to infection with *Bact. pullorum* acquired after the experiment started. This indicates that transmission of *Bact. pullorum* infection among adult fowls by association of infected and non-infected may be an important factor in increasing the extent of the infection in breeding flocks.

Results of Postmortem Examinations of the Fowls in Group 2 that Died

Twenty-four hens died during the year. None of these had given a positive reaction to an agglutination test. Three of the dead were not examined. Twenty-one were given a careful postmortem examination for gross lesions, particularly of the ovary, that might be suggestive of *Bact. pullorum* infection. Cultures were made from the ovaries of these birds.

The bacteriologic examination of eighteen birds gave negative results. The ovaries of eleven of these birds were normal in appearance; a small cyst was attached to the ovaries of two; the ovaries of two birds were congested; and a few small abnormal yolks were present in three birds.

Bact. pullorum was isolated from the ovaries of the remaining three of the twenty-one birds examined. Abnormal yolks were present in all three birds. Two of these had been negative to three agglutination tests and one to five tests. This is additional evidence that fowls with infection of the ovaries with *Bact. pullorum* may fail to react to the agglutination test.

Summary of the Results Obtained with Group 3

This group consisted of fifty fowls that did not react to the first test and were kept separated from all other fowls during the year.

Forty-nine of the birds did not react to any of the agglutination tests during the year. Twelve of these birds died during the year. The deaths occurred after they had been tested from two to ten times. Examination was not made in the case of six of these. Postmortem examination of the remaining six showed the ovaries to be normal in appearance with the exception of a pea-sized tumor-like mass of tissue attached to the ovary of one bird. The bacteriologic examination of these six fowls gave negative results.

One fowl reacted to the fifth and sixth tests. This fowl died soon after the sixth test. The ovary contained several large abnormal yolks from which *Bact. pullorum* was isolated. It seems unlikely that this bird became infected from association with the other birds of group 3 since the experiment started as the results of the agglutination tests and the postmortem examination of those that died indicate that none of the other birds were infected. Furthermore, the lesions, found in

this fowl seemed too extensive to have resulted from recently acquired infection. It appears probable, therefore, that although this fowl did not give a positive reaction until the fifth month of the experiment, it was harboring infection when the experiment started.

Variation in the Degree of the Positive Reactions to the Agglutination Tests

In preceding pages, it has been pointed out that reactions to the agglutination tests of many of the birds fluctuated between positive and negative. A reaction was considered to be positive when there was entire or partial clearing of any one of the four serum-antigen dilutions. It was found that a great variation existed between the degree of the positive reactions of the birds that reacted more than once. In fact, the blood serum of no bird that reacted to more than one test caused the same degree of agglutination in all of the tests in which a positive reaction was secured. The total number of positive reactions obtained was 418. The variation in the degree of reactions is summarized in table 5.

TABLE 5

Variation in the Degree of the 418 Positive Reactions to the Agglutination Test

Dilutions in which agglutination occurred*	Number of reactions	Per cent of total reactions
Partial in 1–25 dilution. No agglutination in others	26	6.1
Complete in 1–25 dilution. No agglutination in others	72	17.2
Partial or complete in 1–25 and 1–50 dilutions. No agglutination in others	133	29.4
Partial or complete in 1–25, 1–50 and 1–100 dilutions. No agglutination in others	71	16.9
Partial or complete in 1–25, 1–50, 1–100 and 1–200 dilutions	116	25.3

* In every instance, there was agglutination in all dilutions below the highest dilution in which agglutination occurred.

It is shown by table 5 that fewer positive reactions would have been obtained if the lowest serum-antigen dilution had been higher than 1–25. If the lowest dilution had been 1–50 there would have been 98 or 23.4 per cent less positive reactions, if the lowest dilution had been 1–100, there would have been 231 or 55.2 per cent less positive reactions.

Such variation in the results of the agglutination tests gives rise to the question of whether an agglutination in low dilution only can be interpreted as indicating infection with *Bact. pullorum*. Some information on this point is furnished by the positive reactions to the agglutination test of seventeen reacting fowls which died and from which *Bact. pullorum* was isolated. Fifty-nine positive reactions were obtained from these seventeen birds. Table 6 gives a summary of the variation in the degree of the reactions.

TABLE 6

VARIATION IN THE DEGREE OF 59 POSITIVE REACTIONS TO THE AGGLUTINATION TESTS OF 17 FOWLS THAT DIED AND FROM WHICH *Bact. pullorum* WAS ISOLATED

Dilutions in which agglutination occurred	Number of reactions	Per cent of total reactions
Partial in 1–25 dilution. No agglutination in others	3	5.0
Complete in 1–25 dilution. No agglutination in others	13	22.0
Partial or complete in 1–25 and 1–50 dilutions. No agglutination in others	20	33.8
Partial or complete in 1–25, 1–50 and 1–100 dilutions. No agglutination in others	11	18.6
Partial or complete in 1–25, 1–50, 1–100 and 1–200 dilutions	12	20.3

By comparing table 6 with table 5, it is seen that the variation in the degree of the positive reaction to the agglutination tests of the seventeen fowls that were known to harbor *Bact. pullorum* closely follows the variation in the degree of the positive reactions of all of the birds. The fact that in more than half of the positive reactions of these seventeen known infected fowls agglutination was obtained only in one or both of the 1–25 and 1–50 dilutions indicates that any fowl that gives a positive agglutination reaction in these dilutions but not in higher dilutions may be a carrier of *Bact. pullorum*. It would, therefore, be expected that any agglutination test procedure for the detection of carriers of *Bact. pullorum* should include a serum-antigen dilution as low as 1–25.

Conclusions

This paper presents the results of the first twelve of a series of at least twenty-four monthly agglutination tests of the same fowls for the detection of *Bact. pullorum* infection, together with the results of the bacteriologic examinations of the fowls that have died during the twelve month period. Complete interpretation of the results of these tests cannot be made until the experiment is terminated and a postmortem and bacteriologic examination is made of all of the fowls. The information obtained from the results of the first year of the experiment, however, would seem to warrant the following conclusions:

Adult fowls with well-established ovarian infection with *Bact. pullorum* may not always react to an agglutination test. This factor seriously affects the dependability of the agglutination test as a means of detecting *Bact. pullorum* carriers and therefore detracts from the practical value of the tests as a means for the complete eradication of the infection from a breeding flock.

A fowl that has reacted to an agglutination test may not react to subsequent tests even though it is still infected with *Bact. pullorum*. Therefore, a fowl that has once reacted to a test cannot be considered as free from the infection if it fails to react to tests that are made subsequently.

A positive reaction to the agglutination test may be considered as a highly accurate indication of *Bact. pullorum* infection. A negative reaction to a test, however, appears to less accurately indicate freedom from *Bact. pullorum* infection, either recently acquired or of long standing.

In an agglutination test procedure with an antigen of equal or greater density than that used in these studies, a serum-antigen dilution at least as low as 1–25 should be included. Clearing of the 1–25 dilution alone or accompanied by clearing of one or more higher dilutions of the same serum can be interpreted as a positive reaction.

No information regarding the interpretation of proagglutination or paradoxical reactions was obtained in these studies since this phenomenon was not encountered.

LITERATURE CITED

[1] Rettger, L. F.
1900. Fatal septicemia among young chicks. New York Med. Jour. 71: 803–805.

[2] ———
1901. Fatal septicemia in young chicks. New York Med. Jour. 73: 267–268.

[3] Rettger, L. F., and S. C. Harvey
1908. Fatal septicemia in young chickens or "white diarrhea." Jour. Med. Res. 18:277–290.

[4] Rettger, L. F.
1909. Further studies on fatal septicemia in young chickens, or "white diarrhea." Jour. Med. Res. 21:115–123.

[5] Rettger, L. F., and F. H. Stoneburn
1909. Bacillary white diarrhea of young chicks. Storrs (Connecticut) Agr. Exp. Sta. Bul. 60:1–57, figs. 1–7.

[6] ———
1911. Bacillary white diarrhea of young chicks (second report). Storrs (Connecticut) Agr. Exp. Sta. Bul. 68:279–301, fig. 1–7.

[7] Rettger, L. F., W. F. Kirkpatrick and F. H. Stoneburn
1912. Bacillary white diarrhea of young chicks (third report). Storrs (Connecticut) Agr. Exp. Sta. Bul. 74:153–185, figs. 1–8.

[8] Rettger, L. F.
1914. Ovarian infection in the domestic fowl and direct transmission of disease to offspring. Jour. Exp. Med. 19:552–561.

[9] Rettger, L. F., W. F. Kirkpatrick, and R. E. Jones
1914. Bacillary white diarrhea of young chicks (fourth report). Storrs (Connecticut) Agr. Exp. Sta. Bul. 77:263–309, figs. 1–4.

[10] Jones, F. S.
1910. Fatal septicemia or bacillary white diarrhea in young chickens. New York, State Vet. Col. Ann. Rpt. 1909–10, pp. 111–129.

[11] ———
1911. Further studies on bacillary white diarrhea in young chickens. New York State Vet. Col. Ann. Rpt. 1910–11, pp. 69–88.

[12] Gage, G. E.
1911. Notes on ovarian infection with *Bacterium pullorum* (Rettger) in the domestic fowl. Jour. Med. Res. 24:491–496.

[13] Jones, F. S.
1913. The value of the macroscopic agglutination test in detecting fowls that are harboring *Bacterium pullorum*. Jour. Med. Res. 27: 481–495.

[14] Beach, B. A., J. G. Halpin, and C. E. Lampman
1927. Results of white diarrhea investigation. Jour. Amer. Vet. Med. Assn. 70:597–605.

THE ELIMINATION OF CLOUDY REACTIONS BY THE USE OF FORMALIN AS A PRESERVATIVE FOR *BACTERIUM PULLORUM* ANTIGEN

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Introduction

All who have used the agglutination test for the detection of fowls that harbor *Bacterium pullorum* have observed the occurrence of excessive turbidity in many tubes which seriously interfered with accurate reading of the reactions. Hitchner,[3] in 1923, reported that the turbidity resulted from the precipitation of fat that is present in the blood serum of some fowls and that it could be avoided by starving fowls for thirty-six hours before blood samples were drawn. Matthews,[4] in 1926, reported studies which he believed demonstrated that such turbidity was due to the presence of a protein rather than a fatty substance in blood serum of fowls. He stated that this protein substance was soluble in weak alkali solution and that clouding of agglutination tests could be avoided by adding a small amount of sodium hydroxide solution to antigen.

Bushnell, Hinshaw and Payne,[5] in 1926, published a very complete discussion of bacillary white diarrhea in fowls which included a résumé of the methods used by various agricultural experiment station laboratories in making agglutination tests. This résumé shows that in twenty-four of twenty-eight laboratories, phenol is used for preservation of the antigen. The amount of phenol used is 0.5 per cent in nineteen laboratories and 0.4, 0.3, 0.25 and 0.2 per cent phenol in one each of four other laboratories. One laboratory was reported as

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[3] Hitchner, E. R. The macroscopic agglutination test as influenced by the fatty content of the blood serum of fowls. Jour. Amer. Vet. Med. Assn. 63: 759–763. 1923.

[4] Matthews, F. P. Obscured reactions in the agglutination test for bacillary white diarrhea. Jour. Immunology 11:499–504. 1926.

[5] Bushnell, L. D., W. R. Hinshaw, and L. F. Payne. Bacillary white diarrhea in fowl. Kansas Agr. Exp. Sta. Tech. Bul. 21:1–858, figs. 1–4. 1926.

using either 0.5 per cent phenol or 0.5 per cent formalin; one as using a coal tar disinfectant (no percentage given); and two as using no preservative in the antigen. It is seen from the above that preservation of antigen by the addition of 0.5 per cent phenol is the prevailing practice. In a further discussion of their own technique and that of other laboratories, these writers disclose that the reason for using 0.3 per cent or a lesser amount of phenol in antigen is to avoid confusing turbidity. The writers also state that "Formolized antigens do not cause a precipitation of the fat-like substance but antigens so prepared are not as reliable as when preserved with phenol." Tittsler, in Pennsylvania, however, is reported by them as using 0.5 per cent formalin for preserving antigen.

The laboratory of the Division of Veterinary Science, California Agricultural Experiment Station, was one of those using antigen containing 0.5 per cent phenol in routine testing of breeding flocks. Difficulty in interpreting the results of the tests on account of turbidity produced by certain sera was frequently encountered. An investigation of means of avoiding turbid or cloudy reactions was, therefore, undertaken. This investigation consisted of a comparison of the results of agglutination tests of the same sera with antigens containing varying amounts of phenol or formalin.

Tests with Antigens Containing 0.1, 0.25, or 0.5 Per Cent Phenol

The antigens were prepared by washing off the growth from 48-hour agar cultures of *Bact. pullorum* with a small amount of physiological salt solution containing 0.5 per cent phenol. For the tests, this was diluted with sufficient physiological saline with or without the addition of phenol to give a reading of 3.5 cm. with a Gates[6] nephelometer and to make the final product contain the desired amount of phenol.

Forty-eight sera were tested with three antigens containing 0.1, 0.25 and 0.5 per cent of phenol respectively. Serum-antigen dilutions of 1–25 and 1–50 were used. Readings were made after 24 hours at 37.5° C and 24 hours at room temperature. The results are as follows:

No positive reactions to the test occurred.

In seventeen of the tests with 0.5 per cent phenolized antigen, there was either increased cloudiness of the fluid or sediment at the bottom of the tubes from the precipitation of a substance from the sera.

[6] Gates, F. L. A method of standardizing bacterial suspensions. Jour. Exp. Med. 31:105–114. 1920.

In ten of the tests with the 0.25 per cent phenolized antigen, there was some increase in cloudiness of the fluid due to a substance in the sera. The cloudiness in these cases was not nearly as marked as that which occurred in the tests of the same sera with 0.5 per cent phenolized antigen.

In none of the tests with the 0.1 per cent phenolized antigen was there any cloudiness or sediment due to precipitation of a substance in the serum. In all 48 tests with this antigen, however, there was either an increase in cloudiness or there was sediment at the bottom of the tubes resulting from the multiplication of contaminating organisms that were present in the sera.

The results of these preliminary agglutination tests of fowl serum with phenolized antigens indicated that cloudiness due to the precipitation of a substance in the serum could be lessened in intensity or entirely avoided by using less than 0.5 per cent phenol in the antigen. When 0.25 per cent phenol was used, the degree of cloudiness from this cause was greatly reduced but not entirely eliminated. When 0.1 per cent phenol was used, no cloudiness from the precipitation of substances in the serum occurred. This latter amount of phenol, however, was insufficient to prevent the multiplication of the contaminating organisms in the serum and, therefore, would be unsatisfactory for use unless the blood was drawn and the test carried out under aseptic conditions.

Tests with Antigens Containing 0.5 Per Cent Phenol, 0.5 Phenol, 0.5 Per Cent Formalin or 0.1 Per Cent Formalin

The 0.5 per cent phenolized antigen and 0.5 per cent formolized antigen were prepared by washing 48-hour agar cultures with a small amount of 0.5 per cent phenolized or formolized physiological salt solution and diluting this concentrated suspension sufficiently with 0.5 per cent phenolized or formolized saline solution at the time of use. The 0.1 per cent formolized antigen was prepared by diluting the concentrated 0.5 per cent formolized antigen with physiological salt solution and 0.1 per cent formolized physiological salt solution at the time of use. The turbidity standard of all antigens was a 3.5 cm. reading with a Gates' nephelometer.

Agglutination tests of 970 sera were made with each of the three antigens. The dilutions of serum and antigen used were 1–25 and 1–50. The results are as follows:

In no case was there any cloudiness or sediment resulting from bacterial multiplication.

Cloudiness occurred in 340, or 35.0 per cent of the tests with phenolized antigen. No cloudiness occurred in the tests with formolized antigens.

A positive reaction in the 0.1 per cent formolized antigen was obtained with 168, or 17.3 per cent of the sera. Of these 168 sera that gave a positive reaction with the 0.1 per cent formolized antigen, 127 sera (or 13.0 per cent of the total sera) also reacted positively with the other two antigens; 13 sera (1.3 per cent of the total sera) also reacted positively with the 0.5 per cent phenolized antigen but not with the 0.5 per cent formolized antigen; 19 sera (1.9 per cent of the total sera) also gave a positive reaction with the 0.5 per cent formolized antigen but not with the phenolized antigen; and 9 sera (0.9 per cent of the total sera) did not react with either of the other two antigens.

All sera that reacted with either the phenolized or 0.5 per cent formolized antigen also reacted with the 0.1 per cent formolized antigen.

Twenty-two of the twenty-eight sera that gave a positive agglutination with the 0.1 per cent formolized antigen and no recognizable agglutination with the phenolized antigen caused clouding of the phenolized antigen. It is possible, therefore, that these twenty-two sera did cause an agglutination of the phenolized antigen which was obscured by the cloudiness. This may account for much of the discrepancy in the results obtained in these tests with the phenolized and 0.1 per cent formolized antigens.

The explanation of the failure of twenty-two of the sera that reacted with the 0.1 per cent formolized antigen to react with the 0.5 per cent formolized antigen, however, is not so apparent. Since the only variable factor was the amount of formalin in the antigens, it seems possible that, in these instances, the 0.5 per cent formalin may have exerted an unfavorable influence on the agglutination of the organisms in the antigen.

The results of these comparative agglutination tests suggested that formalized antigens are more suitable for tests of fowl serum than phenolized antigens. Of the two amounts of formalin used in antigen, i.e., 0.1 per cent and 0.5 per cent, the former seemed more satisfactory. Therefore, additional comparative tests of fowl sera with 0.5 per cent phenolized antigen and 0.1 per cent formolized antigen were carried out.

Tests with Antigens Containing 0.5 Per Cent Phenol or 0.1 Per Cent Formalin

These tests were carried out as opportunity was afforded between February 23 and December 30, 1926, with blood samples from thirty-four flocks.

The methods of preparation and standardization of the antigens were the same as in the preceding tests. Four serum-antigen dilutions, 1–25, 1–50, 1–100, and 1–200, were used in approximately one-third of the tests and two dilutions, 1–25 and 1–50, in the remainder.

Duplicate tests of 4322 sera with two antigens containing 0.5 per cent phenol and 0.1 per cent formalin, respectively, were made. The results are given in table 1.

As shown in table 1, the number of the 4322 sera that reacted with either one or both of the 0.5 per cent phenolized antigen and the 0.1 per cent formolized antigen was 1009 or 23.3 per cent. Of this number, 83 did not react with the phenolized antigen and 41 did not react with the formolized antigen, leaving 885 (20.4 per cent of all tests or 87.7 per cent of all positive tests) that reacted with both antigens.

Cloudiness of the phenolized antigen was caused by 1700 or 39.3 per cent of the sera. The formolized antigen was not affected. The agglutination reaction of 298 of these sera was recorded as positive with both antigens, of 34 as positive with phenolized antigen only, and of 64 as positive with formolized antigen only. By comparing these numbers with the total number of sera that caused agglutination reaction with only one antigen, it is seen that 34 of 41 sera that gave a reaction recorded as positive with phenolized antigen only, and 64 of 83 sera that gave a reaction recorded as positive with formolized antigen only also caused cloudiness of the phenolized antigen.

Since cloudiness of serum-antigen mixtures makes interpretation of agglutination reactions uncertain, it is possible that incorrect readings were made of many or all of the reactions with the phenolized antigen of those sera that caused cloudiness of the phenolized antigen and an agglutination reaction recorded as positive with one antigen only. In such a case, an incorrect interpretation of the agglutination-test reactions with phenolized antigen may have been made of 34 of the 41 sera that were recorded as reacting with phenolized antigen only and of 64 of the 83 sera that were recorded as reacting with formolized antigen only. This would leave but 26 or 0.6 per cent of all tests in which failure to secure the same interpretation of the agglutination reactions with both antigens might not have been due to the real reaction with the phenolized antigen being obscured by cloudiness.

TABLE 1

RESULTS OF AGGLUTINATION TESTS OF 4,322 FOWL SERA WITH 0.5 PER CENT PHENOLIZED ANTIGEN AND 0.1 PER CENT FORMOLIZED ANTIGEN

Number of sera	Number that reacted				Number cloudy with phenolized antigen			
	With both antigens	With phenolized antigen only	With formolized antigen only	Total	Total	Also reacted with both antigens	Also reacted with phenolized antigen only	Also reacted with formolized antigen only
200	3	0	5	8	151	0	0	4
65	0	0	0	0	10	0	0	0
56	3	0	0	3	46	0	0	0
92	10	0	1	11	27	1	0	0
42	1	0	0	1	25	1	0	0
11	0	0	0	0	0	0	0	0
9	1	0	0	1	1	0	0	0
9	2	0	0	2	0	0	0	0
171	54	0	2	56	52	10	0	2
54	9	0	0	9	3	0	0	0
176	34	0	10	44	61	9	0	6
170	27	0	16	43	87	12	0	14
215	0	0	1	1	56	0	0	1
218	38	0	0	38	49	2	0	0
164	24	0	5	29	86	6	0	5
125	43	0	4	47	11	4	0	4
203	21	0	0	21	103	1	0	0
190	48	1	3	52	63	1	0	2
161	34	0	2	36	68	5	0	1
70	0	0	0	0	22	0	0	0
114	27	0	0	27	21	0	0	0
199	67	0	0	67	64	2	0	0
50	23	0	0	23	7	0	0	0
134	13	0	2	15	48	4	0	0
96	3	0	0	3	62	0	0	0
131	2	0	0	2	82	0	0	0
13	2	0	0	2	0	0	0	0
33	5	0	0	5	0	0	0	0
21	4	0	0	4	12	0	0	0
6	0	0	0	0	0	0	0	0
36	6	0	0	6	10	0	0	0
36	6	0	0	6	0	0	0	0
156	29	0	1	30	31	6	0	1
313	167	30	10	207	275	164	28	8
152	21	2	0	23	16	0	0	0
151	21	0	4	25	1	0	0	1
280	137	8	17	162	150	70	6	15
4322	885	41	83	1009	1700	298	34	64

Seven of the fowls whose blood serum had agglutinated the formolized antigen and had neither clouded nor agglutinated the phenolized antigen were secured for autopsy. The postmortem and bacteriological examinations of two of these birds were negative. Four birds had abnormal yolks in the ovaries. *Bact. pullorum* was isolated from three of these. The seventh bird exhibited no ovarian abnormalities, but a mass of fibrinous exudate was present in the pericardial sac. *Bact. pullorum* was isolated from the exudate. These results demonstrate that at least a part of the fowls that reacted with the formolized antigen only were carriers of *Bact. pullorum.*

Variations in the Degree of the Reactions with the Two Antigens

The preceding discussion of the comparative results of the agglutination tests with 0.5 per cent phenolized and 0.1 per cent formolized antigens has shown that 885 or 20.4 per cent of the sera gave a positive reaction with both antigens. In these tests, partial or complete agglutination in any serum-antigen dilution was considered a positive reaction. This classification, therefore, serves to differentiate the sera which produced no agglutination of either one or both antigens from those which produced some agglutination in one or more dilutions with each antigen, but does not indicate whether agglutination occurred in one or more dilutions or whether the number of dilutions in which agglutination occurred was the same for both antigens. In making an accurate comparison of the results with the two antigens, however, consideration of the degree of agglutination obtained with each antigen must be given. A summary of the data on this point is, therefore, included in this paper.

Two dilutions, i.e., 1–25 and 1–50, were used in 3021 tests and four dilutions, i.e., 1–25, 1–50, 1–100 and 1–200 in 1301 tests. The results are as follows:

Two-dilution Tests.—A positive reaction with both antigens was obtained in 641 tests. In 546, or 85.1 per cent, agglutination occurred in the same dilutions of both antigens as follows:

35 sera agglutinated the 1–25 dilution only.
511 sera agglutinated both the 1–25 and 1–50 dilutions.

In 95, or 14.8 per cent, of the positive tests, agglutination did not occur in the same dilutions of both antigens. The variations in these agglutination reactions were:

40 sera agglutinated the 1–25 dilution only of phenolized antigen and both dilutions of formolized antigen.
55 sera agglutinated both dilutions of phenolized antigen and the 1–25 dilution only of formolized antigen.

There was cloudiness of the phenolized antigen in 57 of the 95 tests. This might have been responsible for much of the difference in the readings of the reactions with the two antigens in these tests.

Four-dilution Tests.—A positive reaction in both antigens was obtained in 244 tests.

In 126, or 51.6 per cent, agglutination occurred in the same dilutions of both antigens as follows:

27 sera agglutinated the 1–25 dilution only.
39 sera agglutinated the 1–25 and 1–50 dilutions only.
8 sera agglutinated the 1–25, 1–50 and 1–100 dilutions.
59 sera agglutinated the 1–25, 1–50, 1–100 and 1–200 dilutions.

In 118, or 48.3 per cent, of the positive tests, the dilutions in which agglutination occurred were not the same for both antigens. The agglutination titre in fourteen of these tests was higher with the phenolized than with the formolized antigen, and in 104 tests the titre was higher with the formolized than with the phenolized antigen. The variations in these agglutination reactions are given in table 2.

TABLE 2

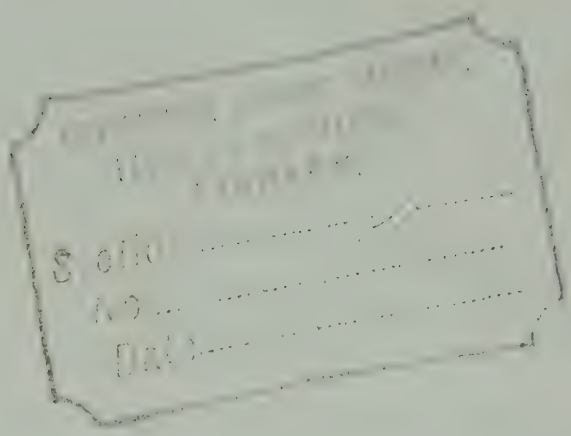
VARIATION IN THE DILUTIONS OF PHENOLIZED AND FORMOLIZED ANTIGENS AGGLUTINATED BY THE SAME SERA. DILUTIONS OF 1–25, 1–50, 1–100 AND 1–200 WERE USED

Number of sera	Dilutions of phenolized antigen agglutinated	Dilutions of formolized antigen agglutinated	Number of sera causing clouding of phenolized antigen
2	1–25, 1–50, 1–100, 1–200	1–25, 1–50	0
3	1–25, 1–50, 1–100, 1–200	1–25, 1–50, 1–100	1
4	1–25, 1–50, 1–100	1–25, 1–50	1
5	1–25, 1–50	1–25	1
6	1–25	1–25, 1–50, 1–100, 1–200	3
28	1–25, 1–50	1–25, 1–50, 1–100, 1–200	6
28	1–25, 1–50, 1–100	1–25, 1–50, 1–100, 1–200	3
14	1–25, 1–50	1–25, 1–50, 1–100	3
5	1–25	1–25, 1–50, 1–100	0
23	1–25	1–25, 1–50	9

It can be seen from the preceding data that there was little difference in the agglutination of the phenolized and formolized antigens in the 1–25 and 1–50 dilutions. Much of the difference that did exist might have been due to incorrect reading of the reaction in the phenolized antigen because of clouding of that antigen by some of the sera. A considerably larger number of sera, however, caused agglutination of the 1–100 and 1-200 dilutions of formolized antigen than in the corresponding dilutions of phenolized antigen.

Discussion

The results of the 4322 comparative tests indicate that antigen containing 0.1 per cent formalin is satisfactory for making agglutination tests of blood serum from fowls. There was little difference in either the number or distribution of the sera which reacted with the two antigens. In the tests in which four dilutions were used and in which reactions to both antigens in at least one dilution were obtained, more sera caused agglutination in the 1–100 and 1–200 dilutions of formolized antigen than in the corresponding dilutions of phenolized antigen.

The cloudiness which occurred in 1700 tests with phenolized antigen did not appear in the corresponding tests with formolized antigen. In this respect, the formolized antigen was more satisfactory than the phenolized antigen.

It was observed that the clumps of bacteria formed by the agglutination of the organisms in the formolized antigen were smaller and more easily broken up than the clumps of bacteria in the phenolized antigen. This was of no importance when complete agglutination occurred, but did make the reading of partial agglutinations more difficult in the formolized antigen than in the phenolized antigen. This feature of the behavior of formolized antigen, however, is an unimportant source of error in the interpretation of agglutination reactions when compared with the frequently-occurring cloudiness of phenolized antigen.